Technical Report

APPROVED: 15 March 2024
doi: 10.2903/sp.efsa.2024.EN-8737

# Technical and scientific assistance on the internal review under Regulation (EC) No 1367/2006 of Commission Implementing Regulation (EU) 2023/2660 renewing the approval of the active substance glyphosate in accordance with Regulation (EC) No 1107/2009

European Food Safety Authority (EFSA)

European Chemicals Agency (ECHA)

## Abstract

Following a joint request of the European Commission, the European Food Safety Authority (EFSA) and the European Chemicals Agency (ECHA) reviewed the scientific arguments raised by non-governmental organisations requesting the review of Commission Implementing Regulation (EU) 2023/2660 renewing the approval of the active substance glyphosate in accordance with Regulation (EC) No 1107/2009. The assessment focuses on relevant scientific elements and does not cover legal aspects, as they are not in EFSA's or ECHA's remit and not in the frame of the mandate received from the European Commission. The current report summarises the outcome of the assessment of the scientific arguments raised by various non-governmental organisations.



**Key words:** glyphosate, peer review, risk assessment, pesticide, herbicide

**Requestor:** European Commission
**Question number:** EFSA-Q-2024-00150
**Correspondence:** pesticides.peerreview@efsa.europa.eu and ECHA Classification classification@echa.europa.eu

**Amendment:** An editorial amendment was carried out that does not materially affect the contents or outcome of this scientific output. For better clarity and to avoid potential confusion, additional explanation has been added when the wording 'Noted' was used in the EFSA responses in Appendix A (pages 80, 92, 102, 105, 107). The original version of the output has been removed from the website, but is available on request.

**Suggested citation:** ECHA (European Chemicals Agency) and EFSA (European Food Safety Authority), 2024. Technical and scientific assistance on the internal review under Regulation (EC) No 1367/2006 of Commission Implementing Regulation (EU) 2023/2660 renewing the approval of the active substance glyphosate in accordance with Regulation (EC) No 1107/2009. EFSA supporting publication 2024:EN-8737. 173 pp. doi:10.2903/sp.efsa.2024.EN-8737

**ISSN:** 2397-8325

## Summary

Following a joint request of the European Commission, the European Food Safety Authority (EFSA) and the European Chemicals Agency (ECHA) reviewed the scientific arguments raised by various non-governmental organisations requesting the review of Commission Implementing Regulation (EU) 2023/2660 renewing the approval of the active substance glyphosate in accordance with Regulation (EC) No 1107/2009. The Commission received five requests submitted by

- Huglo Lepage Avocats ("Huglo Lepage request"),
- Aurelia Stiftung ("Aurelia Stiftung request"),
- Antidote Europe ("Antidote request"),
- Pesticide Action Network Europe, ClientEarth, Future Generation Association, Global 2000, Pesticide Action Network Germany and Pesticide Action Network Netherlands ("PAN request"),
- Secrets toxiques, ANPER, Avenir Sante Environnement, Notre Affaire A Tous, Confederation Paysanne ("Secrets toxiques request")

for an internal review with a view to replacing the re-approval with a decision not to renew the approval of this active substance.

These requests are based on Article 10 of Regulation (EC) No 1367/2006 as amended by Article 1(2)(a) of Regulation (EU) No 2021/1767 of the European Parliament and of the Council of 6 October 2021 amending Regulation (EC) No 1367/2006 on the application of the provisions of the Aarhus Convention on Access to Information, Public Participation in Decision-making and Access to Justice in Environmental Matters to Community institutions and bodies.

EFSA and ECHA analysed the relevant scientific elements put forward in their review requests of the submitting non-governmental organisations and provided jointly the outcome of the assessment of the relevant scientific arguments, referring to the work of the respective agency. In particular ECHA analysed the scientific elements in relation to the harmonized hazard classification carried out by ECHA in accordance with Regulation (EC) No 1272/2008, while EFSA analysed the claims raised with regard to the risk assessment and conclusions derived by EFSA (see Appendix A).

The current report does not cover legal aspects, as they are not in EFSA's or ECHA's remit and not in the frame of the mandate received from the European Commission. Similarly, risk management considerations or matters falling in the remit of risk managers including concerns on the current regulatory system for pesticides were not considered.

# Table of contents

# 1. Introduction

## 1.1. Background and terms of reference as provided by the requestor

Glyphosate is covered under the fifth stage of the renewal work programme (AIR V) in accordance with Regulation (EC) No 844/2012[1].

An application for the renewal of the approval for glyphosate was submitted by a consortium of 8 companies – the Glyphosate Renewal Group (GRG) – and was assessed by four Member States (France, Hungary, the Netherlands and Sweden), appointed to act jointly as rapporteurs for the assessment of the application for renewal of the approval for glyphosate. The four Member States formed the Assessment Group on Glyphosate (AGG) and jointly assumed the role of the rapporteur Member State (RMS).

Following the submission of the renewal assessment report (RAR) to EFSA (received on 15 June 2021), EFSA conducted a peer review of the RAR in line with the provisions of Regulation (EC) No 844/2012. The formal assessment of the proposal for harmonised classification and labelling in accordance with Regulation (EC) No 1272/2008 has been undertaken by the European Chemicals Agency (ECHA) in parallel to the EFSA peer review. When carrying out the risk assessment in the framework of the peer review, EFSA adopted ECHA's hazard assessment and the conclusions of the ECHA Committee for Risk Assessment (RAC) on harmonised classification and labelling delivered in their Opinion on 30 May 2022 (ECHA, 2022). Following the completion of the peer review, including expert discussions, EFSA issued its conclusion on the peer review of the pesticide risk assessment of glyphosate on 6 July 2023 (EFSA, 2023).

The conclusions were reached following the evaluations carried out on the basis of the representative uses of glyphosate as a herbicide, covering uses as pre-sowing, pre-planting and pre-emergence plus post-harvest in vegetables and sugar beet; post-emergence of weeds in orchards, vineyards, row vegetables, railway tracks against emerged annual, biennial and perennial weeds. Moreover, uses as spot treatment against invasive species in agricultural and non-agricultural areas, and in vegetables and sugar beet against couch grass were also included in the EU peer review.

Following delivery of the EFSA Conclusion to the European Commission, in the Standing Committee on Plants, Animals, Food and Feed (SCoPAFF), discussions by risk managers on a decision on the possible renewal of the approval of the active substance took place between September – November 2023, leading to the decision to renew the approval of glyphosate as laid down in Commission Implementing Regulation (EU) 2023/2660[2] of 28 November 2023.

Between 22 December 2023 and 24 January 2024, the Commission received five requests from various non-governmental organisations[3] for internal review of Commission Implementing

[1] Commission Implementing Regulation (EU) No 844/2012 of 18 September 2012 setting out the provisions necessary for the implementation of the renewal procedure for active substances, as provided for in Regulation (EC) No 1107/2009 of the European Parliament and of the Council concerning the placing of plant protection products on the market. OJ L 252, 19.9.2012, p. 26–32.

[2] Commission Implementing Regulation (EU) 2023/2660 of 28 November 2023 renewing the approval of the active substance glyphosate in accordance with Regulation (EC) No 1107/2009 of the European Parliament and of the Council and amending Commission Implementing Regulation (EU) No 540/2011. OJ L, 2023/2660, 29.11.2023.

[3] submitted by
- Huglo Lepage Avocats ("Huglo Lepage request"),
- Aurelia Stiftung ("Aurelia Stiftung request"),
- Antidote Europe ("Antidote request"),

Regulation (EU) 2023/2660 of 28 November 2023 renewing the approval of the active substance glyphosate in accordance with Regulation (EC) No 1107/2009[4] of the European Parliament and of the Council concerning the placing of plant protection products on the market, and amending the Annex to Commission Implementing Regulation (EU) No 540/2011[5].

These requests, submitted by the non-governmental organisations under Article 10 of Regulation (EC) No 1367/2006[6] as amended by Article 1(2)(a) of Regulation 2021/1767[7] of the European Parliament and of the Council of 6 October 2021 amending Regulation (EC) No 1367/2006 on the application of the provisions of the Aarhus Convention, are substantiated by technical arguments referring to EFSA and ECHA's outputs delivered during the procedure for the renewal of approval of glyphosate.

In order to address the issues raised by the non-governmental organisations, EFSA and ECHA was asked for an in-depth analysis of the relevant scientific elements included in the requests for internal review related to:

1. paragraphs 31 to 201 of the request made by PAN;
2. paragraphs 25 to 41 of the request made by Secrets toxiques;
3. from page 23 up to the end of the request made by Aurelia Stiftung; and
4. any part of the requests made by Antidote and Huglo Lepage, referring to the work of the respective agency.

## 2. Assessment

EFSA and ECHA analysed the relevant scientific elements put forward in their review requests of the submitting non-governmental organisations and provided jointly the outcome of the assessment of the relevant scientific arguments, referring to the work of the respective agency.

In particular ECHA analysed the scientific elements in relation to the assessment of the proposal for harmonized hazard classification in accordance with Regulation (EC) No 1272/2008 while EFSA analysed the claims raised with regard to the risk assessment and conclusions derived by EFSA.

The current report does not cover legal aspects, as they are not in EFSA's or ECHA's remit and not in the frame of the mandate received from the European Commission. Similarly, risk management considerations or matters falling in the remit of risk managers including concerns on the current regulatory system for pesticides were not considered.

- Pesticide Action Network Europe, ClientEarth, Future Generation Association, Global 2000, Pesticide Action Network Germany and Pesticide Action Network Netherlands ("PAN request")
- Secrets toxiques, ANPER, Avenir Sante Environnement, Notre Affaire A Tous, Confederation Paysanne ("Secrets toxiques request")

[4] Regulation (EC) No 1107/2009 of the European Parliament and of the Council of 21 October 2009 concerning the placing of plant protection products on the market and repealing Council Directives 79/117/EEC and 91/414/EEC. OJ L 309, 24.11.2009, p. 1–50.

[5] Commission Implementing Regulation (EU) No 540/2011 of 25 May 2011 implementing Regulation (EC) No 1107/2009 of the European Parliament and of the Council as regards the list of approved active substances. OJ L 153, 11.6.2011, p. 1–186.

[6] Regulation (EC) No 1367/2006 of the European Parliament and of the Council of 6 September 2006 on the application of the provisions of the Aarhus Convention on Access to Information, Public Participation in Decision-making and Access to Justice in Environmental Matters to Community institutions and bodies. OJ L 264, 25.9.2006, p. 13–19.

[7] Regulation (EU) 2021/1767 of the European Parliament and of the Council of 6 October 2021 amending Regulation (EC) No 1367/2006 on the application of the provisions of the Aarhus Convention on Access to Information, Public Participation in Decision-making and Access to Justice in Environmental Matters to Community institutions and bodies. OJ L 356, 8.10.2021, p. 1–7.

The relevant scientific arguments and the corresponding scientific views and replies by EFSA and ECHA on the specific points raised are presented in the format of a reporting table for each internal review request.

The arguments raised are summarised in column 2 of the reporting tables. The scientific views and conclusions by EFSA and ECHA are outlined in column 3 of the tables.

The finalised reporting tables for each internal review request are provided in Appendix A of this report.

# Documentation provided to EFSA and ECHA

1. Letter from the European Commission to the EFSA and ECHA Executive Directors, dated 1 March 2024 requesting technical and scientific assistance on the internal review under Regulation (EC) No 1367/2006 of Commission Implementing Regulation (EU) 2023/2660 of 28 November 2023 renewing the approval of the active substance glyphosate in accordance with Regulation (EC) No 1107/2009.

2. Pesticide Action Network Europe, ClientEarth, Future Generation Association, Global 2000, Pesticide Action Network Germany and Pesticide Action Network Netherlands ("PAN request") for internal review regarding Commission Implementing Regulation (EU) 2023/2660 of 28 November 2023 renewing the approval of the active substance glyphosate.

3. Huglo Lepage Avocats ("Huglo Lepage request") for internal review regarding Commission Implementing Regulation (EU) 2023/2660 of 28 November 2023 renewing the approval of the active substance glyphosate.

4. Aurelia Stiftung ("Aurelia Stiftung request") for internal review regarding Commission Implementing Regulation (EU) 2023/2660 of 28 November 2023 renewing the approval of the active substance glyphosate.

5. Antidote Europe ("Antidote request") for internal review regarding Commission Implementing Regulation (EU) 2023/2660 of 28 November 2023 renewing the approval of the active substance glyphosate.

6. Secrets toxiques, ANPER, Avenir Sante Environnement, Notre Affaire A Tous, Confederation Paysanne ("Secrets toxiques request") for internal review regarding Commission Implementing Regulation (EU) 2023/2660 of 28 November 2023 renewing the approval of the active substance glyphosate.

# References

AGG (Assessment Group on Glyphosate), 2021. Renewal Assessment Report (RAR)/ CLH report on glyphosate prepared by the four rapporteur Member States of AGG, consisting of FR, HU, NL and SE, in the framework of Commission Implementing Regulation (EU) No 844/2012. Available online: www.efsa.europa.eu and Registry of CLH intentions until outcome - ECHA (europa.eu)

AGG (Assessment Group on Glyphosate), 2023. Revised Renewal Assessment Report (RAR) on glyphosate prepared by the four rapporteur Member States of AGG, consisting of FR, HU, NL and SE, in the framework of Commission Implementing Regulation (EU) No 844/2012, May 2023. Available online: www.efsa.europa.eu

Debode F, Caulier S, Demeter S, Dubois B, Gelhay V, HulinJ, Muhovski Y, Ninane V, Rousseau G, and Bragard C, 2024. Roadmap for the integration of environmental microbiomes in risk assessments under EFSA's remit. EFSA supporting publication 2024:EN-8602.93pp. doi:10.2903/sp.efsa.2024.EN-8602

ECHA and EFSA (European Chemicals Agency and European Food Safety Authority with the technical support ofthe Joint Research Centre (JRC)), Andersson N, Arena M, Auteri D, Barmaz S, Grignard E, Kienzler A, Lepper P,Lostia AM, Munn S, Parra Morte JM, Pellizzato F, Tarazona J, Terron A and Van der Linden S, 2018. Guidance for the identification of endocrine disruptors in the context of Regulations (EU) No 528/2012 and (EC) No1107/2009. EFSA Journal 2018;16(6):5311, 135 pp. https://doi.org/10.2903/j.efsa.2018.5311

ECHA (European Chemicals Agency), 2022. Committee for Risk Assessment (RAC) Opinion proposing harmonisedclassification and labelling at EU level of glyphosate (ISO); N-(phosphonomethyl)glycine. CLH-O-0000007122-85-01/F. Available online:www.echa.europa.eu.

ECHA (European Chemicals Agency), 2022. Response to a specific question from ENVI Committee exchange of views. Document reference D(2022)0887. Available online: Glyphosate - ECHA (europa.eu)

ECHA (European Chemicals Agency), 2022. Response to the report from Health and Environment Alliance (HEAL). Available online: Glyphosate - ECHA (europa.eu).

ECHA (European Chemicals Agency), 2023. Response from ECHA and EFSA to the European Commission related to letter from NGOs. Document reference Ref. D(2023)0927. Available online: Glyphosate - ECHA (europa.eu).

EFSA (European Food Safety Authority), 2009. Guidance on Risk Assessment for Birds and Mammals on request from EFSA. EFSA Journal 2009;7(12):1438, 358 pp. doi:10.2903/j.efsa.2009.1438

EFSA (European Food Safety Authority), 2011. Submission of scientific peer-reviewed open literature for the approval of pesticide active substances under Regulation (EC) No 1107/2009. EFSA Journal 2011;9(2):2092, 49 pp. doi:10.2903/j.efsa.2011.2092

EFSA (European Food Safety Authority), 2014. Guidance on the assessment of exposure of operators, workers, residents and bystanders in risk assessment for plant protection products. EFSA Journal 2014;12(10):3874, 55pp.https://doi.org/10.2903/j.efsa.2014.3874

EFSA (European Food Safety Authority), Arena M, Auteri D, Barmaz S, Brancato A,Brocca D, Bura L, Carrasco Cabrera L, Chiusolo A, Civitella C, Court Marques D, Crivellente F,Ctverackova L, De Lentdecker C, Egsmose M, Erdos Z, Fait G, Ferreira L, Greco L, Ippolito A, Istace F,Jarrah S, Kardassi D, Leuschner R, Lostia A, Lythgo C, Magrans JO, Medina P, Mineo D, Miron I, Molnar T,Padovani L, Parra Morte JM, Pedersen R, Reich H, Sacchi A, Santos M, Serafimova R, Sharp R, Stanek A,Streissl F, Sturma J, Szentes C, Tarazona J, Terron A, Theobald A, Vagenende B, Van Dijk J and Villamar-Bouza L, 2018. Conclusion on the peer review of the pesticide risk assessment of the active substance cypermethrin. EFSA Journal 2018;16(8):5402, 28 pp. https://doi.org/10.2903/j.efsa.2018.5402

EFSA (European Food Safety Authority), 2019. Statement on the available outcomes of the human health assessment in the context of the pesticides peer review of the active substance chlorpyrifos. EFSA Journal 2019;17(8):5809, 23 pp. https://doi.org/10.2903/j.efsa.2019.5809

EFSA (European Food Safety Authority), Ippolito A, Focks A, Rundlöf M, Arcella A, Marchesi M, Neri FM, Szentes Cs, Rortais A and Auteri D, 2021. Analysis of background variability of honeybee colony size. EFSA supporting publication 2021:EN-6518. 79pp. doi:10.2903/sp.efsa.2021.EN-6518, available at https://efsa.onlinelibrary.wiley.com/doi/abs/10.2903/sp.efsa.2021.EN-6518

EFSA (European Food Safety Authority), Auteri D, Arce A, Ingels B, Marchesi M, Neri FM, Rundlöf M, Wassenberg J, 2022. Analysis of the evidence to support the definition of Specific Protection Goals for bumble bees and solitary bees. EFSA supporting publication 2022:EN-7125. 68 pp.doi:10.2903/sp.efsa.2022.EN-7125I, available at https://efsa.onlinelibrary.wiley.com/doi/abs/10.2903/sp.efsa.2022.EN-7125

EFSA (European Food Safety Authority), Alvarez F, Arena M, Auteri D, BinagliaM, Castoldi AF, Chiusolo A, Colagiorgi A, Colas M, Crivellente F, De Lentdecker C, Egsmose M, Fait G,Ferilli F, Gouliarmou V, Herrero Nogareda L, Ippolito A, Istace F, Jarrah S, Kardassi D, Kienzler A,Lanzoni A, Lava R, Leuschner R, Linguadoca A, Lythgo C, Magrans O, Mangas I, Miron I, Molnar T,Padovani L, Parra Morte JM, Serafimova R, Sharp R, Szentes C, Terron A, Theobald A, Tiramani M andVillamar-Bouza L, 2022. Conclusion on the peer review of the pesticide risk assessment of the activesubstance isoflucypram. EFSA Journal 2022;20(6):7328, 34 pp. https://doi.org/10.2903/j.efsa.2022.7328

EFSA (European Food Safety Authority), Alvarez F, Arena M, Auteri D, Binaglia M,Castoldi AF, Chiusolo A, Colagiorgi A, Colas M, Crivellente F, De Lentdecker C, Egsmose M, Fait G, FerilliF, Gouliarmou V, Herrero Nogareda L, Ippolito A, Istace F, Jarrah S, Kardassi D, Kienzler A, Lanzoni A,Lava R, Leuschner R, Linguadoca A, Lythgo C, Magrans O, Mangas I, Miron I, Molnar T, Padovani L,Parra Morte JM, Rizzuto S, Serafimova R, Sharp R, Szentes C, Terron A, Theobald A, Tiramani M and Villamar-Bouza L, 2022. Conclusion on the peer review of the pesticide risk assessment of the active substance benfluralin. EFSA Journal 2022;20(9):7556, 28 pp. https://doi.org/10.2903/j.efsa.2022.7556

EFSA (European Food Safety Authority),Alvarez, F., Arena, M., Auteri, D.,Binaglia, M., Castoldi, A. F., Chiusolo, A., Crivellente, F., Egsmose, M., Fait, G., Ferilli, F., Gouliarmou, V.,Nogareda, L. H., Ippolito, A., Istace, F., Jarrah, S., Kardassi, D., Kienzler, A., Lanzoni, A.,...Villamar-Bouza, L. (2023). Peer review of the pesticide risk assessment of the active substance glyphosate. EFSA Journal, 21(7),1–52, https://doi.org/10.2903/j.efsa.2023.8164

EFSA (European Food Safety Authority), Adriaanse P, Arce A, Focks A, Ingels B,J ¨olli D, Lambin S, Rundl ¨of M, S ¨ußenbach D, Del Aguila M, Ercolano V, Ferilli F, Ippolito A, Szentes Cs,Neri FM, Padovani L, Rortais A, Wassenberg J and Auteri D, 2023. Revised guidance on the riskassessment of plant protection products on bees (Apis mellifera,Bombusspp. and solitary bees). EFSAJournal 2023;21(5):7989, 133 pp. https://doi.org/10.2903/j.efsa.2023.7989

EFSA PPR Panel (EFSA Panel on Plant Protection Products and their Residues), 2010; Scientific Opinion on the development of specific protection goal options for environmental risk assessment of pesticides, in particular in relation to the revision of the Guidance Documents on Aquatic and Terrestrial Ecotoxicology (SANCO/3268/2001 and SANCO/10329/2002). EFSA Journal 2010;8(10):1821. [55 pp.] doi:10.2903/j.efsa.2010.1821. https://www.efsa.europa.eu/en/efsajournal/pub/1821

EFSA PPR Panel (EFSA Panel on Plant Protection Products and their Residues), 2013. Guidance on tiered risk assessment for plant protection products for aquatic organisms in edge-of-field surface waters. EFSA Journal 2013;11(7):3290, 186 pp. doi:10.2903/j.efsa.2013.3290

EFSA PPR Panel (EFSA Panel on Plant Protection Products and their Residues), 2014. Scientific Opinion addressing the state of the science on risk assessment of plant protection products for non-target terrestrial plants. EFSA Journal 2014;12(7):3800, 163 pp. doi:10.2903/j.efsa.2014.3800, available at https://www.efsa.europa.eu/en/efsajournal/pub/3800

EFSA PPR Panel (EFSA Panel on Plant Protection Products and their Residues), 2015. Scientific Opinion addressing the state of the science on risk assessment of plant protection products for non-target arthropods. EFSA Journal 2015;13(2):3996, 212 pp. doi:10.2903/j.efsa.2015.3996 available at https://efsa.onlinelibrary.wiley.com/doi/abs/10.2903/j.efsa.2015.3996

EFSA Scientific Committee, 2016. Guidance to develop specific protection goals options for environmental risk assessment at EFSA, in relation to biodiversity and ecosystem services. EFSA Journal 2016;14(6):4499, 50 pp. doi:10.2903/j.efsa.2016.4499 https://efsa.onlinelibrary.wiley.com/doi/epdf/10.2903/j.efsa.2016.4499.

EFSA Scientific Committee, 2011. Scientific Opinion on genotoxicity testing strategies applicable to food and feed safety assessment. EFSA Journal 2011;9(9):2379, 69 pp. https://doi.org/10.2903/j.efsa.2011.2379.

EFSA Scientific Committee, Hardy A, Benford D, Halldorsson T, Jeger MJ,Knutsen HK, More S, Naegeli H, Noteborn H, Ockleford C, Ricci A, Rychen G, Schlatter JR, Silano V,Solecki R, Turck D, Benfenati E, Chaudhry QM, Craig P, Frampton G, Greiner M, Hart A, Hogstrand C,Lambre C, Luttik R, Makowski D, Siani A, Wahlstroem H, Aguilera J, Dorne J-L, Fernandez Dumont A,Hempen M, Valtue~na Martınez S, Martino L, Smeraldi C, Terron A, Georgiadis N and Younes M, 2017. Scientific Opinion on the guidance on the use of the weight of evidence approach in scientific assessments. EFSA Journal 2017;15(8):4971, 69 pp. https://doi.org/10.2903/j.efsa.2017.4971

EFSA Scientific Committee, 2017. Scientific Opinion on the clarification of some aspects related to genotoxicity assessment. EFSA Journal 2017;15(12):5113, 25 pp. https://doi.org/10.2903/j.efsa.2017.5113.

EFSA Scientific Committee, 2021. Guidance on aneugenicity assessment. EFSA Journal 2021;19(8):6770, 27 pp. https://doi.org/10.2903/j.efsa.2021.6770

European Commission, 2002. Guidance Document on Terrestrial Ecotoxicology Under Council Directive 91/414/EEC. SANCO/10329/2002-rev. 2 final, 17 October 2002

European Commission, 2014. Assessing potential for movement of active substances and their metabolites to ground water in the EU. Report of the FOCUS Workgroup. EC Document Reference SANCO/13144/2010-v. 3, 613 pp, as outlined in Generic guidance for tier 1 FOCUS groundwater assessment, v. 2.2 May 2014

Haines-Young R and Potschin MB, 2018. Common International Classification of Ecosystem Services (CICES) V5.1 and Guidance on the Application of the Revised Structure. Available from www.cices.eu

Merten C, Schoonjans R, Di Gioia D, Pelaez C, Sanz Y, Maurici D, Robinson T, 2020. Editorial: Exploring the need to include microbiomes into EFSA's scientific assessments. EFSA Journal 2020;18(6):e18061, 7 pp. https://doi.org/10.2903/j.efsa.2020.e18061

Moreno, Francisco Javier; Florencio Pazos, Manuel Garrido-Romero, Cyrielle Payen, Gonzalo Borrego-Yaniz, Mónica Chagoyen, Nieves Corzo, Martine Denis, Christelle Fablet, María Fernández, Adela Granja, Maryse Guinebretière, Muriel Guyard, Rodrigo Jiménez-Saiz, Alassane Keita, Annaëlle Kerouanton, Ana Márquez, Javier Martín, Antonia Montilla, Ana Muñoz-Labrador, Jorge Novoa, Frédéric Paboeuf, Marta G. Rivera-Ferre, Patricia Ruas-Madiedo, Lorena Ruiz, Amandine Thépault, Mar Villamiel, Carlos Benito and Marianne Chemaly, 2024. Roadmap for the integration of gastro-intestinal (GI) tract microbiomes (human and domestic animal) in risk assessments under EFSA's remit. EFSA supporting publication 2024:EN-8597. 238 pp. doi:10.2903/sp.efsa.2024.EN-8597

# Abbreviations

| | |
|---|---|
| ADI | acceptable daily intake |
| ALS | amyotrophic lateral sclerosis |
| a.i. | active ingredient |
| AOEL | acceptable operator exposure level |
| ARfD | acute reference dose |
| a.s. | active substance |
| ASD | autism spectrum disorder |
| CLH | harmonized classification and labelling |
| CLP | classification, labelling and packaging |
| DNA | deoxyribonucleic acid |
| DNT | developmental neurotoxicity |
| ECHA | European Chemicals Agency |
| EPA | Environmental Protection Agency of the United States |
| EQS | environmental quality standards |
| GBHs | glyphosate-based herbicides |
| GC-MS | gas chromatography–mass spectrometry |
| GD | guidance |
| GIS | Geographic Information System |
| GLY | glyphosate |
| HQ | hazard quotient |
| IARC | International Agency for Research on Cancer |
| IPA | isopropylamine salt of glyphosate |
| LOAEL | lowest observed adverse effect level |
| LOD | limit of detection |
| $LR_{50}$ | lethal rate, median |
| MIE | molecular initiating event |
| MS | Member State |
| NHL | non-Hodgkin lymphoma |
| NOAEL | no observed adverse effect level |
| NOEC | no observed effect concentration |
| OECD | Organisation for Economic Co-operation and Development |
| PD | Parkinson's disease |
| PND | post-natal day |
| PPP | plant protection product |
| RAC | regulatory acceptable concentration; also stands for ECHA's Risk Assessment Committee |

| | |
|---|---|
| RAR | Renewal Assessment Report |
| REACH | Registration, Evaluation, Authorisation of Chemicals Regulation |
| RMS | Rapporteur Member State |
| SCoPAFF | Standing Committee on Plants, Animals, Food and Feed |
| SPG | Specific Protection Goal |
| SVL | snout–vent length |
| TG | test guideline |
| TEU | Treaty of the European Union |
| TMS | trimesium |
| TRVs | toxicological reference values |
| US EPA | United States Environmental Protection Agency |
| WoE | weight of evidence |

# Appendix A – Collation of the relevant scientific arguments provided in the review requests for the active substance glyphosate and the conclusions drawn by EFSA and ECHA on the specific points raised

**Relevant scientific arguments provided in the review letter submitted by PAN & others for the active substance glyphosate and the conclusions drawn by ECHA on the specific points raised**

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | Paragraph(s) (unless more specific reference mentioned) | The quoted text from the relevant paragraphs are from the machine translations of the original text in French (where applicable) | Notes: Throughout, references to the CLH report also include its annexes, including the DAR volumes |
| 1 | Paragraphs 39-41 (as relevant) | Paragraph 40: "These results are qualified because of the existence of a 'negative result for induction of DNA repair (UDS)'[59]. That argument relating to the existence of a negative UDS test (for 'unscheduled DNA synthesis') has been repeated for more than 20 years during each assessment of glyphosate, both for its classification under CLP Regulation No 1272/2008 and for its approval as an active substance. However, this type of test was removed from the OECD TGs in 2014 due to its unreliability." | In relation to the UDS assay, the RAC opinion actually states that "*Two negative Unscheduled DNS Synthesis (UDS) assays using primary hepatocytes was presented in the CLH dossier (CA 5.4.1/033, 1994; CA 5.4.1/034, 1984). The studies were considered to be not acceptable by the DS due to deviations from the OECD TG 482. RAC notes that the UDS assay is no longer a standard method and that the OECD TG 482 has been deleted*".<br><br>The conclusion in the RAC opinion (P42) on the results from 2 in vivo Comet assays (Bolognesi et al. (1997) and Mañas et al. (2013)) was that "*These two studies suggest that glyphosate may induce increases in DNA strand breaks that are rapidly repaired following a single exposure. That glyphosate may induce increases in DNA strand breaks is supported by the in vitro Comet assays, but the data also appear to show that the increases in strand breaks reach a plateau with no further increase with increasing dose. The biological significance of a slight increase in DNA strand breaks as reported following exposure to glyphosate in the drinking water is uncertain*". |
| 2 | Paragraph 54 (last sentence) and paragraph 56 | "it is pungent to note that in its opinion on the carcinogenicity of glyphosate, ECHA itself acknowledges doubts as to the genotoxic nature of | ECHA has published their response to this issue (published on the ECHA website at Microsoft Word - D(2022)0887_MR2109.docx (europa.eu)), as follows: |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | glyphosate, deploring the absence of an in vivo comet test and the examination of tissues other than spinal mole[82]".<br><br>The footnote (82) refers to the following text from P 48 of the RAC opinion: "*Glyphosate appears to induce transient DNA strand breaks as observed in the in vitro and in vivo Comet assays or by using the alkaline elution assay; however, no reliable in vivo Comet assays were included in the CLH dossier in relevant target organs e.g., liver and kidney or a TGR somatic and germ cell gene mutation assay. There is also some evidence that glyphosate may induce oxidative stress in certain cells and tissues with the potential to induce oxidative DNA-lesions that may lead to mutations if not repaired. However, the gene mutation assays were all negative and bone marrow mutagenicity was considered negative in a weight of evidence assessment of the available oral and i.p. micronucleus assays. Noting the absence of a Comet assay conducted according to OECD TG 489 in relevant tissues as well as a TGR somatic and germ cell gene mutation assay conducted according to OECD TG 488, the biological importance of such DNA lesions in relation to mutagenicity is equivocal*". | "Firstly, RAC is obliged under CLP to classify on the basis of the available information and evaluations are always carried to a conclusion in the categories specified by the regulation.<br><br>Having said this, and as noted in the opinion, according to the criteria in the CLP Regulation, classification as Category 2, is largely based on positive evidence obtained from somatic cell mutagenicity tests in mammals or other in vivo somatic cell genotoxicity tests which are supported by positive results from in vitro mutagenicity assays. The gene mutation assays in the data assessed were all negative and bone marrow mutagenicity was considered negative in a weight of evidence assessment of the available oral micronucleus assays and intraperitoneal micronucleus assays.<br><br>The statement quoted from the opinion related to the Comet assay and Transgenic rodent (TGR) somatic and germ cell gene mutation assays which are two particular assays among many other lines of evidence potentially informing a classification. The opinion noted the absence of these assays/studies in relevant tissues, but also noted that the biological importance of such DNA lesions (i.e., as identified from these assays) in relation to mutagenicity is equivocal, therefore the fact that some studies of this type were not included is not crucial for the conclusion.<br><br>More specifically, the data available for evaluation of germ cell mutagenicity is extensive and includes studies covering bacterial and mammalian cell in vitro mutagenicity assays as well as in vivo mammalian mutagenicity assays and even some human data. Furthermore, according to the opinion, the data includes studies of sufficient reliability and relevance to allow a robust evaluation, especially in the perspective of the requirements of the CLP Regulation. In RAC's view, the data were sufficient to arrive at a robust conclusion without these assays/studies". |
| 3 | Paragraph 66 | "The EFSA conclusions on the absence of carcinogenicity of glyphosate are based on and are in line with ECHA's | ECHA rejects the assertions made in this paragraph. Detailed responses are provided below. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | 2022 opinion on the classification and labelling of glyphosate[93]. However, this view is vitiated by manifest errors of assessment. It is also based on analyses that do not comply with OECD guidelines and ECHA's own guidance documents, in breach of the principles of excellence and independence". | |
| 4 | Paragraph 67 | "These points are developed in detail by two independent experts in Annex 4". | Additional points raised in Annex 4 are addressed separately under the relevant headings in Annex 4, below. |
| 5 | Paragraph 67 | Minimisation of the results of carcinogenicity of glyphosate in the studies provided by the applicants for re-approval | ECHA rejects the assertions made in this paragraph. Detailed responses are provided below. |
| 6 | Paragraph 69 (reproduced in full) | "Had these flaws been avoided, the results of the regulatory studies should have led to the finding that there is sufficient evidence of carcinogenicity of glyphosate and thus to the conclusion of an assumed carcinogenic potential for humans (category 1B classification) (based on CLP, Annex I, 3.6.2.2.3 (b), in particular the statement) "*a causal relationship has been established between the agent and an increased incidence of malignant neoplasms or of an appropriate combination of benign and malignant neoplasms in (….) (b) two or more independent studies in one species carried out at different times or in different laboratories or under different protocols*[94]. It is apparent from an objective and rigorous analysis of the regulatory studies submitted by the applicants for re-approval that the second condition is manifestly satisfied". | ECHA has responded to this argument in their responses to a report from HEAL, which has been published on the ECHA website since July 2022 and can be accessed from 40ee075a-8b57-f524-9a82-b492a77a53f1 (europa.eu).<br>This argumentation ignores the obligation from the CLP Regulation to weigh all of the available evidence in each case. In Recital 33, of the CLP Regulation, this is reflected as follows: "*Recognising that the application of the criteria for the different hazard classes to information is not always straightforward and simple, manufacturers, importers and downstream users should apply weight of evidence determinations involving expert judgement to arrive at adequate results*." The following provisions are also relevant to this: Article 9(3) of the CLP Regulation; Section 1.2 of Annex XI to the REACH Regulation and CLP Annex I (section 1.1.1.3 ) (both referred to in Art 9(3) of the CLP Regulation);<br><br>A weight of evidence assessment means that data is given different weight depending on factors such as the quality and consistency of the results. Also, in relation to the statement in the CLP Regulation that "both positive and negative results shall be assembled together in a single weight of evidence determination" (emphasis added), this is not a matter of a majority of studies supporting one or the other outcome. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | | Thus, RAC is obliged to make an overall weight of evidence analysis of the complete data set. In the case of glyphosate, some studies were found to be of no weight, and were not included in the analysis. For example, two studies in mice which were negative for carcinogenicity were considered to be conducted with too low doses and "*did not comply with current standards*" (CA 5.5/022, 1988 and Report no. 80 10; CA 5.5/024, 1982 original report, revised 1992) and, therefore, were considered as unacceptable.<br><br>In addition to multiple animal studies, data from the epidemiology studies and genotoxicity studies were also considered in the weight of evidence assessment. RAC concluded that despite some indications of carcinogenicity seen in some studies mainly in mice, the criteria for classification are not met when all the studies and findings are considered together. Thus, RAC reached the conclusion that no classification for carcinogenicity is warranted. |
| 7 | 70-82 | Findings of keratoacanthomas are relevant for classification | ECHA rejects the assertions made in this paragraph. Detailed responses are provided below. |
| 8 | 71-78 | Use of biased statistical methods | ECHA rejects the assertions made in this paragraph. Detailed responses are provided below. |
| 9 | 71 | ECHA uses two strategies to disregard conclusions resulting from a trend analysis. | There was (and is) no strategy to disregard conclusions from a trend analysis. The various statistical analyses (including trend analysis) are presented in the RAC opinion for each tumour type examined as part of the assessment of the findings.<br><br>Some of the outcomes arising from the statistical analyses are dependent on which test is used.<br>• None of the tumours referred to in the PAN Request for internal review were statistically significant using pairwise comparison with two-sided testing<br>• Some were statistically significant following two-sided testing using the trend test<br>• Additional findings were also statistically significant when one-sided pairwise comparisons or trend tests were employed.<br>These considerations were included in the assessment. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| 10 | 72 | Pairwise vs trend test: [ECHA] "considers that those findings are not necessarily corroborated by pair comparisons".<br><br>"this reasoning is contrary to the OECD Guidelines"<br><br>"These guidelines also point out that, according to the guidelines of the United States Environment Protection Agency (hereinafter referred to as the EPA), trend studies and pair comparisons are alternative methods of establishing statistical significance: ` *Significance in either kind of test is sufficient to reflect the hypothesis that chance accounts for the result*'[98]". | It is a fact that the findings are not necessarily corroborated by pair-wise comparisons.<br><br>References here are to the OECD Guidance document 116. Both types of tests (trend and pair-wise testing) are described in the document, including some of the benefits and disadvantages of each approach.<br><br>The CLH report as well as the Opinion included the results for both one- and two-sided testing from both trend as well as pairwise comparisons of the incidences of tumours. In the CLH report, the data from the Portier (2020) paper for the one-sided testing from both types of tests was included alongside the 2-sided data in the tables describing each tumour type. The fact that (according to OECD 116) a trend test is more powerful was taken into account in the analysis. The opinion stated that "*RAC notes that the analysis made by Crump et al. (2020, B.6.5.18.1) shows that statistically significant effects on tumour incidences should be carefully evaluated for biological relevance due to the high number of studies assessed, as chance findings may occur.*" (p65, 72). The point was elaborated on in some detail in the CLH report, and summarized in the opinion as follows: "*Due to the IARC conclusion, experts have investigated why there are different conclusions from different investigating bodies (Crump et al., 2020, B.6.5.18.1; Portier et al., 2020, B.6.5.18.2).*<br><br>*Crump et al. (2020, B.6.5.18.1) pointed out that the animal carcinogenicity data on glyphosate are extensive (≥ 15 long term rodent oral bioassays of glyphosate identified by US EPA (2016), EFSA (2016) and IARC (2015). Each bioassay was conducted in both sexes, with each sex potentially having 40-60 unique tumour types, resulting in over 1000 potential statistical tests, which could result in many statistically significant ($p \leq 0.05$) tumours by chance alone – approximately 5%. Crump et al. (2020, B.6.5.18.1) further assessed the probability of false positives using a modification of the permutation approach of Farrar and Crump (1988 and 1990). The analysis made by Crump et al.* |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | | *(2020, B.6.5.18.1) showed that statistically significant effects on tumour incidences should be carefully evaluated for biological relevance as chance findings may occur.*<br><br>*Portier et al. (2020, B.6.5.18.2) also provided an additional revised statistical evaluation and trend test analyses of relevant tumour types reported in the carcinogenicity studies but did not take into account the chance effect due to multiple testing as pointed out by Crump et al. (2020, B.6.5.18.1). Furthermore, as indicated in the OECD Guidance document 116, statistical significance is only part of the interpretation of the biological importance of a particular finding. In the CLH dossier, as well as in the current RAC assessment, the tumour types showing statistically significant trends in the analysis by Portier et al. (2020, B.6.5.18.2) were taken into consideration in the assessment of cancer types. One of the differences between the study by Portier et al. (2020, B.6.5.18.2) and the analysis by the DS was that Portier used 1-sided testing with a significance level of 0.05, whereas in the original study reports and the DS analysis 2-sided testing was applied with a significance level of 0.05 (which is equivalent to 1-sided testing using a significance level of 0.025)"* (P48-49 of the opinion).<br><br>Therefore, the analysis of the statistical testing was part of the thorough analysis of the data conducted by RAC. Statistical analyses formed one part of the RAC assessment for biological significance.<br><br>RAC also noted that when using trend tests, significant trends are in some cases related to smaller increases in tumours only reported in the high dose group with no or low incidences in the control group. In these cases, provided the findings were not significant in pairwise testing, the strength of the evidence was considered to be weak.<br><br>There is no indication in the guidance document that only one or the other type of statistical analysis should exclusively be used (see also below). An objective analysis should not favour using a methodology which drives towards a pre- |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | | determined conclusion, but should consider all the relevant factors. Hence although RAC considered the results from Trend tests in their analysis, RAC did not exclusively rely on these results. The analyses conducted were consistent with the requirement in CLP in relation to applying a weight of evidence approach (Annex I, 1.1.1.3 and Article 9(3)), as well as OECD GD 116.<br><br>The text from the paragraph from OECD GD 116 quoted at the end of paragraph 72 continues as follows: "*A statistically significant response may or may not be biologically significant and vice versa. The selection of a significance level is a policy choice based on a trade-off between the risks of false positives and false negatives. A significance level of greater or less than 5% (the most common significance level) is examined to see if it confirms other scientific information. When the assessment departs from a simple 5% level, this should be highlighted in the risk characterization. A two-tailed test or a one-tailed test may be used. In either case a rationale is provided*. " The focus was on determining whether each finding was biologically significant. |
| 11 | 73-77<br><br>(1) Paragraph 73, last sentence<br>(2) Paragraph 74, 1st and 2nd sentences<br>(3) Paragraph 75<br>(4) Paragraph 77 2nd sentence onwards | Unilateral (one- sided) vs bilateral (2-sided) tests<br><br>(1) "As a matter of principle, it is twice as difficult to establish the statistically significant character of a scenario through a bilateral test (the probability that chance is responsible for statistical correlation should be less than 0.025) than through a one-sided test (the probability threshold is 0.05)."<br><br>(2) "There is in principle no sense to use a bilateral pair comparison test to | According to OECD GD 116 (§ 384, p 133), reproduced here in full "*In a carcinogenicity study, the expectation is often that the change will be an increase in tumours in the treated group so a one-sided test may be considered more appropriate, although this can be controversial. If the treatment could also be protective (i.e., reduce tumour incidence or delay it) then a two-sided comparison may be more appropriate. Regulatory authorities may have specific opinions. For instance, the US EPA (2005) notes that either "a two tailed test or a one-tailed test may be used*" (emphasis added). Therefore, in accordance with GD 116 it is also acceptable to conduct either one or two tailed tests.<br><br>As quoted in the Request for internal review from PAN, OECD 116 states that '*The statistical methods most appropriate for the analysis of results, given the experimental design and objectives, should be established before starting the study*'. This establishes the starting point for the |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | assess the carcinogenicity of glyphosate". Indeed, it is clear that glyphosate does not have a protective effect against cancer, so it would be entirely fanciful to take into account the purely theoretical scenario of a combination of the substance with a decrease in cancer cases".<br><br>(3) "Against all expectations, however, ECHA – and the RMS in the RAR – used a bilateral test, following which they concluded that the role of chance in increasing tumour impacts could not be entirely excluded"<br><br>(4) "It should be noted that, according to the OECD guidelines, 'The statistical methods most appropriate for the analysis of results, given the expert design and objectives, should be established before starting the study'[100]. This means that it is up to the designers of the study not merely to indicate the statistical method used, but to establish its appropriateness. However, neither the ECHA opinion nor the RAR mentions – let alone discuss – the | analyses: The method chosen by the parties compiling the original study report.<br><br>The approach to statistical analyses by the dossier submitter (DS) was explained on p 257 of the CLH report, as follows:<br><br>"The statistical analyses provided by AGG are based on values reported in the original study reports, the statistical re-assessment of the data given in the previous CLH report (2016) and/or by AGG own statistical analysis. However, both one- or two-sided significance can be calculated, depending on the hypothesis to test. OECD Guidance Document 116 stipulates "*The choice of whether to use a one- or two-sided test should be made at the design rather than the analysis stage. A two-sided statistical hypothesis test tests for a difference from the negative control (in a pairwise comparison) in either direction. A one-sided comparison tests for a difference in only one pre-specified direction, but as a consequence has more power. In a carcinogenicity study, the expectation is often that the change will be an increase in tumours in the treated group so a one-sided test may be considered more appropriate, although this can be controversial. If the treatment could also be protective (i.e., reduce tumour incidence or delay it) then a two-sided comparison may be more appropriate*". In the AGG overall analysis on the tumour relevance, two-sided testing was applied as this is in line with how the statistical analysis was established in the study protocols of the available carcinogenicity studies".<br><br>The RAC opinion also transparently summarised the approach to statistical analyses used "*The main statistical methods used in the animal studies were the Fisher's exact test for pairwise comparisons and the Cochran-Armitage trend test, and in this opinion these two methods are referred to unless stated otherwise. In their detailed assessment of findings, the DS repeated both the pairwise and trend test statistical calculations for the findings from relevant studies (eight studies in rats and five studies in mice; for details, see below). In addition, for one study in mice (CA 5.5/016, 2001), a Peto-analysis was* |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | justifications provided by applicants for re-approval in support of the choice of a bilateral test, most probably because no justification is presented in the study protocols (which the Commission can easily verify in the context of the present request for review). As a result, in clear breach of the standards of excellence, transparency and independence, the authorities have shown blind confidence in the choice of the statistical method used by applicants for re-approval, even though (i) the method chosen is in principle the least suitable and not the one recommended by the OECD; and that (ii) this method, in that it makes it twice as difficult to detect a statistically significant association, is clearly in favour of the interests of those applicants. | *performed for the induction of malignant lymphomas*." (P 48, under "Summary of the Dossier Submitters Proposal")<br><br>Therefore, contrary to the statement in the PAN request for internal review, the DS and RAC did not merely consider the statistical analyses conducted in the study report, but as indicated above (response to Paragraph 72), transparently compiled and took into account the various alternative approaches to statistical analysis for each tumour type, including those indicated in the publication by Portier (2020). This clearly does not reflect blind confidence in the choice of statistical methods as claimed.<br><br>In relation to statements (1) and (2) from Paragraphs 73 and 74, ECHA notes that the goal is for an objective analysis of the significance of the tumour incidences observed.<br><br>Concerning (3) and (4), the specific tests used are those described in OECD GD 116 under the heading 4.15 "Standard (simple) statistical analysis of qualitative data", which include the Fisher exact test and the Cochran-Armitage trend test. The Peto test was used in one (2001) study in mice. As explained in response to the points above, the DS and RAC went beyond what was included in the original study reports by also providing results from additional testing of the data (including from 1 and 2 sided testing using the standard trend tests and pairwise testing referred to above).<br><br>The primary information used in developing the RAC opinion is that from the CLH report as well as the comments received during the Consultation of the CLH report. From the comments received during the Consultation, it is clear that not all statisticians agree with the assertion that since glyphosate is not a protective treatment against cancer, the use of the two-sided statistical test is incomprehensible, because one party (a statistician, in Comment 21 of the "Response to comments document (RCOM), accessible from COMMENTS AND RESPONSE TO COMMENTS ON CLH: PROPOSAL AND JUSTIFICATION (europa.eu)) noted that they had |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | | concerns about the use of one-sided significance levels (exclusively in the direction of a positive association) to summarise the results of the glyphosate rodent studies and that "*the use of one-sided p-values for positive associations will not only increase statistical power, but will also increase the number of false positive findings*". The comment also pointed to the findings of Crump et al (2020), in which it is stated, after analysing ten of the rodent carcinogenicity studies for positive and negative dose-response trends using the same statistical trend test, that they found [marginally] more evidence for negative dose-response trends than positive i.e., as expressed in the comment "*more tumor types showing significant decreases in tumor rates with increasing glyphosate levels than there were showing significant increases*". The focus of the analyses in the CLH report as well as RAC was on the findings indicating increased tumours, and these were analysed in detail. Taking into account whether there are increasing or decreasing trends in tumour incidences is also part of this analysis, to ensure that potential false positive as well as false negative findings are appropriately addressed. |
| 12 | 79-82 | Paragraph 79: "79. ECHA also seeks to reduce the relevance of statistical analyses by invoking arguments in a manner contrary to the OECD guidelines and its own guidance document." | ECHA rejects the assertion that it has sought to minimize the relevance of statistical analyses by invoking arguments in a manner contrary to the OECD guidelines and its own guidance document. |
| 13 | 80-81 | (1) Paragraph 80: … "the weight of evidence of those analyses is reduced in so far as tumours were observed only in male rats, and not in females or mice"<br>(2) Paragraph 81: "It follows, therefore, that while a concordance of results between the sexes and species is such as to increase the weight of the observations, | (1)- (2) The opinion at pages 92/93 concluded that "*There is insufficient evidence to support a classification in Category 2 based on the evaluation of seven rat studies*". Furthermore, it is stated that "*The conclusion is supported by the benign nature of the tumours with no suggestions of progression towards malignancy, a low strength of the evidence and a lack of consistency between sexes and across the many studies performed*". The evidence was simply not sufficiently strong to conclude that classification was warranted and the evidence was certainly not sufficient to indicate that there was a sex-specific tumour. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | observations relating to a single sex of a single species may already be sufficient to demonstrate the carcinogenicity of a substance." | |
| 14 | 82 | (3) "ECHA minimises the scope of the observations of Keratoacanthomas in male rats on the ground that such tumours are common in that species of rats. This is based on a single study that is supposed to provide a reliable historical monitoring database. In the absence of a match in terms of year and laboratory, those data cannot be regarded as reliable"<br><br>(4) "In addition, the use of historical monitoring data is marginalised in the applicable guidelines. On the one hand, the OECD guidelines referred to above emphasise that data from the concurrent monitoring group, i.e. the study itself (which does not reveal a common trend towards the development of | (3) ECHA notes that these tumours were not previously discussed in the EU and the discussion in the context of the RAC opinion was based on this having been raised in the publication by Porter et al (2020).<br>The skin keratoacanthoma observations in the studies referred to by Portier et al (2020) were not statistically significant by either pairwise comparison or by trend test (two-sided testing), but Portier et al (2020) found a statistically significant trend following one-sided trend testing. No skin keratoacanthoma findings were seen in two studies. RAC noted in the opinion (P64) that skin keratoacanthoma is a benign tumour which is shown to be rather common in aged male rats and in fact were only reported in male rats (and not in female rats nor in male or female mice). Furthermore, no malignant squamous cell carcinomas were reported. In humans, this type of benign skin tumour is associated with multiple exposure to sunlight, whereas in rats, which are most likely only exposed to artificial light, the cause of skin keratoacanthomas is unknown. RAC concluded that the increase in skin keratoacanthomas only reported in male rats is not of sufficient relevance for classification for carcinogenicity. A more detailed analysis of the skin keratoacanthoma findings from this and other studies where this was observed is in the RAC opinion (under the heading "Skin tumours").<br><br>The single study referred to (Zwicker et al, 1992) is not cited as historical control data for the tumour type, as claimed (the actual incidences in the study were not referred to either in the opinion or in the CLH report). The point is made that the tumour type is rather common in aged male rats. The article itself notes that (in terms of skin neoplasms in aged SD rats, the topic of the article) "Keratoacanthoma was the most frequent epithelial neoplasm in males". This is an additional factor |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | such tumours) should always be preferred to historical monitoring data" | appropriately taken into account in the assessment and consideration of the timing was part of assessing this consideration. The differences in the incidences in the studies assessed between male and female rats reflect the differences seen in the article by Zwicker et al (1992).<br><br>(4) ECHA agrees that the relevant OECD guidelines correctly emphasise that data from the concurrent control group (i.e. the study itself) should always be preferred to historical control data (HCD). In the opinion, for each finding RAC noted the historical control data, but the emphasis was placed on comparisons with the concurrent control. |
| 15 | 83-88 | Paragraph 83: "ECHA uses similar arguments to reject the findings of three mice studies (out of a total of five) showing a statistically significant increase in kidney tumours in males" | |
| 16 | 84 | (1) "It claims, first, that historical control data on mice do not support those findings. This statement is incorrect for two out of three studies. As for the third, the historical monitoring data are not sufficiently clear to allow any conclusions to be drawn".<br><br>(2) Paragraph 84, second sentence: "as for the third, the historical monitoring data are not sufficiently clear to allow any conclusions to be drawn", | It actually was not stated in the opinion that the HCD data do not support the findings. The opinion noted (P67) that HCD were not available for one study, in one study the findings were slightly above the HCD and in a third study the findings were within the HCD range.<br><br>The basis for this statement by PAN, which presumably refers to the study in Swiss mice is not clear. In relation to this point, the RAC opinion at page 67 states that "*Spontaneous control incidences for Swiss male mice included for the CA 5.5/016 (2001) study were based on eight studies performed between 1996 and 2002, with a mean of 2.0% and a range of 0 - 6%. The increased incidence of renal tubular adenomas in the CA 5.5/016 (2001) study was within the HCD and is therefore considered incidental and not related to glyphosate exposure.*" |
| 17 | 85 | Regarding the arguments against the statement in the opinion that there is "no evidence for a progression | (1) It is not clear what is meant by the statement |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | to malignancy" (paragraph 85, 1st sentence):<br>(1) "This is not only contrary to the OECD Guidelines (which simply require " guidance", not " evidence")"<br><br>(2) Paragraph 85, 2nd sentence: "the potential for progression of this type of tumour from adenoma to the carcinogoma stage is well known. Moreover, the duration of the studies was too short to be able to draw conclusions from the absence of such progression". | It is noted in page 67 of the RAC opinion that "*differentiation between tubular cell adenoma and tubular cell carcinoma is not always clearly apparent and both lesions are derived from the same cell type. Accordingly, it is the combined incidences that have been used in the statistical analysis*." The findings were seriously considered by RAC.<br>The study durations were all within those prescribed in the OECD Guidelines for mice (18 or 24 months) |
| 18 | 86 | "ECHA also excludes results related to high dose administration on the grounds that these doses are too high and exceed the limit dose of 1 g/kg". | RAC actually does not refer to the limit dose in their assessment of carcinogenicity against the CLP criteria.<br>RAC notes at page 67 (in relation to the kidney tumours) that "*In two of the three positive studies (CA 5.5/018-019, 1997; CA 5.5/023, 1983), increased tumour incidences were only observed at very high doses (> 4000 mg/kg bw/d) at which the body weight gain in males were decreased compared to controls by up to 11% and 15% in the CA 5.5/023 (1983) and the CA 5.5/018-019 (1997) study, respectively. The OECD TG 452 for carcinogenicity studies does not give a precise top dose recommendation, but states that the highest dose level should normally be chosen to identify the principal target organs and toxic effects while avoiding suffering, severe toxicity, morbidity, or death, and the highest dose level should be chosen to elicit evidence of toxicity, as evidenced by, for example, depression of body weight gain (approximately 10%). RAC therefore gives less weight to the findings at these very high dose levels. RAC also notes that the mouse is exposed to glyphosate via the diet with a high exposure to the gastrointestinal tract. The human relevance of the renal tumours at very high doses is considered to be low*" …<br>In the RCOM (P38) is the DS noted that the OECD TG 453 states that "*a limit of* |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | | *1000 mg/kg bw/day may apply except when human exposure indicates the need for a higher dose level to be used*". Furthermore, in paragraph 117 of OECD GD 116, it is stated that "*As indicated in the Test Guidelines, a top dose not exceeding 1000 mg/kg body weight/day may apply except when human exposure indicates the need for a higher dose level to be used*". Thus, giving less weight to doses at or greater than 1000 mg/kg bw/day for glyphosate is consistent with the guideline as the exposure to humans is far below this level.<br><br>Contrary to what is claimed by PAN ,RAC did not dismiss the tumour findings at doses above 1000 mg/kg bw/day, but the findings at the very high doses (above 4000 mg/kg bw/d) were given lower weight, for the reasons explained in the Opinion as well as in the CLH report. In short, while there were low incidences of tumours at the highest doses in these studies, this was in combination with other effects (body weight gain data suggesting general toxicity) and therefore RAC, following recommendations in the relevant OECD guideline, gave these findings lower weight amongst all the other information available to inform on carcinogenicity. |
| 19 | 87 | "ECHA further states that a plausible mechanistic explanation for the occurrence of such tumours is missing. However, there are perfectly plausible explanations, including the genotoxic potential of glyphosate (reference made to para. 34 of the document, under genotoxicity) and its ability to produce oxidative stress, a possibility recognised by ECHA itself and demonstrated by independent literature in particular the Gao et al. (2019). This shows how glyphosate produces oxidative stress in the same organ (kidney), species (mice) and sex (male) as those where tumours | This issue has been addressed in a response from ECHA which is published on the ECHA website (accessible from 7e7b03bc-b2f1-8949-b6f6-da9feb1f7292 (europa.eu)) as follows: In the context of the CLP criteria the primary source of evidence to inform on classification is enumeration of tumours in animal studies and determination of their level of statistical significance. Many other factors can be taken into consideration including mode of action/mechanistic considerations. Oxidative stress is a mechanism that can lead to tumour formation and therefore falls into the latter category as a factor that can be taken into consideration when assessing tumour incidences. ECHA's independent assessment is based on a large number of scientific studies designed to examine the hazardous properties of glyphosate, including whether it causes cancer. All available evidence was carefully examined to arrive at a conclusion. No important findings were dismissed. Tumour incidences in the available studies were |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | appeared in carcinogenicity studies". | examined in detail and the conclusion was that there was no convincing evidence that glyphosate induces tumours.<br>In the absence of clear evidence of tumours linked to glyphosate, evidence that glyphosate causes oxidative stress is not relevant for the conclusion. Findings of oxidative stress in a study are not on their own sufficient for classification. In particular, potential mode of action considerations arising from one study cannot provide support in the absence of convincing evidence for carcinogenicity in another study.<br>The mechanistic data from the Gao et al (2019) study were included in the CLH report and considered in the RAC opinion (P43-44 and 74-75). |
| 20 | 89 -95 | "Four out of five mice carcinogenicity studies showed an increasing incidence of cases of malignant lymphoma when exposed to glyphosate. In three of these studies, this growth is statistically significant".<br>"ECHA again dismisses the relevance of these results on the basis of several highly problematic arguments" | Please see below for a detailed response. |
| 21 | 91 | "ECHA again uses a bilateral pair comparison test to challenge the statistical significance observed on the basis of a trend test." | The argument is rejected, as discussed above, in the response to paragraphs 71-78 |
| 22 | 92 | "ECHA considers that the results are not corroborated by historical monitoring data. That statement is simply not correct."<br>Reference: Annex 4 | The usual term in animal studies in this context is "historical control data". This argument relates to the statement in the conclusions to this section that "*the maximum incidences in the majority of the studies were considered to be within the historical control range for the CD-1 mice, although adequate HCD were not available for all studies*". It was noted in the RAC opinion that the tumour incidence of 12% at the high dose of 4348 mg/kg bw/d (a very high dose) in the study by CA 5.5/018-019 (1997) was within the historical control range for Crj:CD-1 male mice obtained from seven studies, since the range was 3.8% to 19.2% with a mean of 7%. The opinion also noted, however, that "*six of the seven studies had a control incidence ≤ 6% leading to a range of 3.8% to 6% with a mean of 4.92%. Therefore,* |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | | *when taking into account HCD from the six studies the incidences of malignant lymphoma in male mice exceeded the HCD*" (see page 71 of the opinion). The HCD status was clearly and accurately reported.<br><br>This is also the case for Swiss albino mice, where it is stated in the opinion that "*In Swiss albino mice (CA 5.5/016, 2001), the incidence of malignant lymphoma in male and female mice at the top dose was 38% and 50%, respectively. However, the high background incidence in this strain must be taken into consideration. The HCD in males had a mean of 15.8% with a range of 6 - 30% and in females a mean of 33% with a range of 14 - 58%. Thus, the incidences of malignant lymphomas were above the upper range of the HCD for the male mice.*" (see page 71 of the opinion). |
| 23 | 93 | ECHA considers that the increases were limited to high doses. Apart from being irrelevant (see above, paragraph 78), this statement is also incorrect since in two studies effects were observed at other doses. | It is assumed that the statement refers to the findings in study CA 5.5/012-015 (2009) and CA 5.5/016 (2001), where the incidences at the intermediate doses were intermediate between those in the controls and the high dose groups. The findings in the 2009 study at the intermediate doses were low (1/51 = 2% at the low dose and 2/51 = 4% at the mid-dose). In the 2001 study, the findings at the low dose and mid-dose were 15/50 = 30% and 16/50 = 32%, respectively, and were observed against a high background in the male controls (10/20 = 50%).<br><br>The statistical significance of malignant lymphomas observed in these studies were noted by the DS to be very much dependent on the statistical method used for analysing the data. In the 2009 study, the findings were statistically significant when the trend test was applied (either one- and two-sided). They were also not statistically significant at the high dose when a pairwise comparison was performed (but were statistically significant with a one-sided pairwise test (Portier, 2020)). The increased incidence in the 2001 study was not confirmed either by the trend test (one- or two-sided) or by a two-sided pairwise test but only when using a one-sided pairwise test and one-sided Peto-analysis. The increased incidence in the 2001 was against a high background incidence. RAC has reviewed all of the data and in a weight of evidence |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | | assessment concludes that the reported incidences of malignant lymphoma in CD-1 mice and Swiss mice is not considered related to glyphosate exposure. A more detailed analysis of the malignant lymphoma findings from these and other studies where this tumour type was observed is in the RAC opinion (under the heading "Malignant lymphoma" at page 69 of the opinion). |
| 24 | 94 | "ECHA observes that lymphoma increases are not consistent from one sex to another. However, as explained above (see paragraph 81 above), it is apparent from ECHA's own guidance document that this cannot preclude a finding of carcinogenicity". | The findings in the 2 sexes were indeed not consistent.<br><br>The incidences of the findings in control females were equivalent to those at the highest dose in males in two studies and more than 2-fold those at the highest dose in the remaining 3 studies. There were no significant findings lymphoma findings in females in any of the studies.<br><br>The reference is to the CLP Guidance from 2015. In the latest version (2017) the relevant section also states (P383-384) that "*Effects seen only in one sex in a test species may be less convincing than effects seen in both sexes, unless there is a clear patho-physiological difference consistent with the mode of action to explain the single sex response. However, there is no requirement for a mechanistic understanding of tumour induction in order to use these findings to support classification*".<br><br>The analysis conducted by RAC was completely consistent with the CLP Guidance. |
| 25 | 95 | "ECHA considers that the possible role of oncogenic viruses cannot be ignored. For the reasons set out in detail in the experts' analysis, this claim is unfounded and is based on the abusive extrapolation of a single article" | The quote is from the summary of the proposal of the dossier submitter. RAC does not refer to the role of oncogenic viruses in its assessment. |
| 26 | 96-97 | Epidemiological studies: PAN quotes from the RAC opinion references to "a weak association can be seen for persons with a relatively high exposure (third tertile) and acute myeloid leukaemia and non-Hodgkin's Lymphoma" and | The quote from p 54 of the RAC opinion is from the summary of the dossier submitter's proposal. It is also taken out of context.<br><br>The relevant paragraph in p. 54 of the opinion is quoted below in full: "*Andreotti et al. (2018) showed that, based on the data from the AHS cohort, no overall* |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | "Overall, available epidemiological case-control studies, reviews, re-analyses and meta-analyses show weak statistically significant associations between exposure to glyphosate-based herbicide and findings of cancer, especially non-Hodgkin's Lymphoma" and argue that these reflect the definition of "limited evidence of carcinogenicity" and therefore should have led to at least classification as Carc. 2. | *association between exposure to glyphosate-based herbicides and cancer was reported. However, a weak association can be seen for persons with a relatively high exposure (third tertile) and acute myeloid leukaemia and non-Hodgkin's Lymphoma after a 20-year lag time (time between exposure and tumour development). These data also concern a very small research population of n=15 and n=8 cases, respectively, and therefore the DS considered these findings to be of questionable value. However, the DS noted the finding of a possible association with acute myeloid leukaemia should be looked at carefully in future updates on the AHS data. The DS, however, noted that a high number of cancer sites were analysed so there was the possibility of statistical findings by chance and that acute myeloid leukaemia was not observed in any of the other epidemiological studies with glyphosate*".<br><br>Following their own assessment, RAC concluded that "*A causal relationship with exposure to glyphosate-based herbicide can thus not be confirmed by RAC. More specifically, this is due to a number of factors – i.a. the weak associations which were only significant when certain statistical tests were applied, small studies with low number of exposed cases, the probability of recall bias for previous exposure (duration and dose) especially in the case-control studies, selection bias, the lack of biomonitoring data, frequently not adjusting for confounding factors such as co-exposure to other pesticides and risk estimates often getting lower when more comprehensive adjustment was applied, the presence of a toxic co-formulant (POE-tallowamine), and the changes in the definitions of non-Hodgkin's Lymphoma/other cancers over the years*". (see page 91 of the opinion)<br><br>The reasons why the conclusions were reached are described in considerable detail in the opinion (and by the DS in the DAR). It was only after careful consideration of all the information that RAC concluded at page 91 of the opinion that "*No association between exposure to glyphosate-based herbicides and incidences of non-Hodgkin's Lymphoma* |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | | *was observed in the only robust cohort study available*".<br>In addition it was indicated in the opinion that "*The findings from the epidemiology studies are used in a weight of evidence approach together with the findings in animal studies*". |
| 27 | 98 | "The biased and manifestly erroneous nature of the assessment stems in particular from the arbitrary and unfounded exclusion of high dose results and the equally arbitrary choice (based solely on the unreasoned choice of applicants for re-approval) of a priori inappropriate statistical analysis method that is at least capable of revealing the carcinogenicity of a substance. It should also be mentioned that ECHA could not rely on any regulatory study considered "reliable without restriction" to assess the carcinogenicity of glyphosate".<br><br>"As regards modes of action, it should also be recalled that ECHA itself acknowledges doubts as to the lack of genotoxicity of glyphosate, deploring the absence of an in vivo comet test and the examination of tissues other than spinal mole (see pt. 52)." | ECHA rejects categorically the claims of "biased and manifestly erroneous nature of the assessment".<br><br>The statistical tests used were all examined openly in the assessment.<br><br>If ECHA cannot rely on any regulatory study considered "reliable without restriction" to assess the carcinogenicity of glyphosate, the information base becomes very limited and would seriously undermine the whole process of regulation of chemical substances and mixtures in the EU (and more broadly).<br><br>As regards the concern that key tests were not conducted due to a statement in the RAC opinion referring to the absence of specific assays in relevant target organs (OECD TG 489 "the comet assay" and OECD TG 488 "TGR"), firstly, it should be noted that the CLH process assesses available data – there is no mechanism to generate additional information.<br><br>Secondly, the statement quoted from the opinion related to the Comet assay and Transgenic rodent (TGR) somatic and germ cell gene mutation assays which are two particular assays among many other lines of evidence potentially informing a classification. The opinion noted the absence of these assays/studies in relevant tissues, but also noted that the biological importance of such DNA lesions (i.e., as identified from these assays) in relation to mutagenicity is equivocal, therefore the fact that some studies of this type were not included is not crucial for the conclusion. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | | Furthermore, the data available for evaluation of germ cell mutagenicity is extensive and includes studies covering bacterial and mammalian cell in vitro mutagenicity assays as well as in vivo mammalian mutagenicity assays and even some human data. Furthermore, according to the opinion, the data includes studies of sufficient reliability and relevance to allow a robust evaluation, especially in the perspective of the requirements of the CLP Regulation. In RAC's view, the data were sufficient to arrive at a robust conclusion without these assays/studies.<br><br>ECHA stands by the assessment conducted and conclusions drawn by the independent scientists in RAC on the hazardous properties of glyphosate. |
| Points raised in Annex 4, submitted with the PAN request to review. | | | |
| 28 | Annex 4 | | As a general point, RAC sets out to conduct an objective analysis to determine whether or not there is a hazard, without a preconceived view as to whether classification is warranted.<br><br>A scientific assessment can have differences in opinion between different groups of scientists. However, RAC is a group of independent scientists. |
| 29 | Section 2.0 "Strength of evidence" | | See below |
| 30 | Page 2, 2nd last paragraph | While numerous statistical methods exist for the comparison of incidences, two different approaches can be distinguished in principal among these methods: pairwise comparisons where incidences in the control group are compared with those of an individual treatment group; and trend tests, which evaluate statistical significance by integrating the incidences in the control and all treated groups. In its flow chart, OECD [6] (p. 123) explicitly recommends trend test while in the same document (p. 116) it is stated "Trend | As explained above (see responses to Paragraphs 72-78) results of trend tests were considered alongside the other approaches to statistical analysis. Ultimately statistical analyses form one part of the analysis of whether any findings are of biological significance. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | tests and pairwise comparison tests are the recommended tests …", and that "Significance in either kind of test is sufficient to reject the hypothesis that chance accounts for the result." And again, it is pointed out: "A statistically significant response may or may not be biologically significant and vice versa."<br><br>From that it can be concluded, that per se there is no basis to disregard the results of trend tests. Thus, any decision to exclude a significant increase in tumours via a trend test because the pairwise tests are not significant has no basis in the guidelines used by ECHA. The same is true when pairwise tests are significant but the trend is not. | |
| 31 | Page 3 | "OECD [6] (p.114) notes that "The statistical methods most appropriate for the analysis of results, given the experimental design and objectives, should be established before commencing the study." (see also [3, 4])<br>Establishing the most appropriate method before commencing the study cannot just mean mentioning what statistical method will be used in the study plan, but instead providing a rationale why a particular statistical method was considered most appropriate. Whether this was done in the case of the glyphosate carcinogenicity studies needs to be demonstrated, because the study plans have not been disclosed. Based on the experience of a toxicologist who worked as a study director in contract research and industry from 1997 – | The specific tests used are those described in OECD GD 116 under the heading 4.15 "*Standard (simple) statistical analysis of qualitative data*", which include the Fisher exact test and the Cochran-Armitage trend test. The Peto test was used in one (2001) study in mice. As explained in response to points above, the DS and RAC went beyond what was included in the original study reports by also providing results from additional testing of the data (including from 1 and 2 sided testing using the standard trend tests and pairwise testing referred to above).<br><br>This response also applies to the paragraph which follows the one quoted. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | 2009, it appears unlikely that such rationales have been provided in the study plans. | |
| 32 | Section 3.0 "Factors that strengthen a statistical finding" | 3.1 Historical Control Data (HCD) | ECHA is of the view that RAC and the DS used the HCD in a manner consistent with the CLP Regulation and the CLP Guidance |
| | | 3.2 Consistency | |
| 33 | | 3.2.1 Consistency across sexes<br><br>"Thus, additional data is required before a determination can be made regarding a single-sex finding and it is the responsibility of the ECHA to obtain this information to determine if single-sex findings are valid. There is no indication in this language that observing a positive response in only one sex automatically results in a reduction in the weight of evidence" | CLP Annex I: 3.6.2.2.5. quoted in full states as follows: "*The factors can be viewed as either increasing or decreasing the level of concern for human carcinogenicity. The relative emphasis accorded to each factor depends upon the amount and coherence of evidence bearing on each. Generally there is a requirement for more complete information to decrease than to increase the level of concern. Additional considerations should be used in evaluating the tumour findings and the other factors in a case-by-case manner*"<br><br>The analyses in the RAC opinion are consistent with the CLP text, including consideration of consistency of findings across studies. RAC did not argue that observing a positive response in only one sex automatically results in a reduction in the weight of evidence. |
| 34 | | 3.2.2 Consistency across strains and species<br><br>However, there is no indication in the guidance that a failure to have the same positive response across multiple species reduces the weight of evidence. There is no discussion about different strains (although one might presume the same arguments could hold) nor any discussion of what constitutes consistency in this situation when there are more than one study using the same strain or different strains of the same species. | The obligation for RAC to use weight of evidence in their assessment is addressed above (response to Paragraph 69, above) |
| 35 | 4.1 Skin Keratoacanthomas | Various arguments are presented under this heading, including the following:<br>4.1.1 Strength of evidence | As explained above, this is a factor in the assessment of all the relevant information. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | "They mention no significant pairwise comparisons, suggesting this weakens the findings from the trend tests in contradiction of their guidelines". (P9) | |
| 36 | | "Doing that makes the incidence counts 1/50, 2/50, 0/50, 0/50, 5/50 from low to high dose. Had the ECHA bothered to do the trend test with these data, they would have noted the one-sided p-value decreases to 0.017 (exact) from 0.047 (as presented in Portier) and the two-sided (approximate) trend test used by ECHA changes to 0.008 from 0.07". (P 9-10) | The incidence counts are exactly as presented in the opinion. RAC presented both 1 and 2 sided trend test results which gave the values published in the opinion. |
| 37 | | "In only one of the four positive studies (CA 5.5/010, 1990 [13]) is the dose-response monotonic non-decreasing; in the remaining three studies [12, 14, 15], the general trend is increasing, but it can go up and then down". (P10) | The authors of the Annex consider the findings to reflect a dose-response relationship even when there is no linear dose-response relationship. RAC considered the findings to be insufficient for classification. Some of the factors was that the increased incidences above controls were only observed at very high doses (>= 940 mg/kg/d) except in one study (out of six) where increases were seen at intermediate doses. In 2 studies no increases were seen at any dose, even despite a high highest dose having been used.<br><br>The opinion:<br>"In four out of the six acceptable rat carcinogenicity studies, increased incidences of skin keratoacanthomas were observed in the high dose group. A dose-response relationship was only reported in one of the studies (CA 5.5/010, 1990)."<br><br>"Overall, RAC considers that depending on the statistical method used, the increased incidences of skin keratoacanthomas in male rats were either non-significant, borderline, or significant. However, RAC notes that when performing trend tests, in cases where effects only occur at the highest dose, it is high dose levels that trigger the statistical significance in a trend test. Skin keratoacanthomas were only reported in male rats and not in female rats or male and female mice. The |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | | incidences exceeded the available HCD range or from individual studies when available, however, noting that the HCD are very limited for the induction of skin keratoacanthomas in male rats. Skin keratoacanthoma is a benign tumour which is shown to be rather common in aged male rats (Zwicker et al., 1992)". (P64 of the opinion). |
| 38 | | "Using an ad-hoc limit, as done here, to eliminate a positive effect would result in many truly carcinogenic substances having no significant trends in test animals and would allow many of these carcinogenic substances back on the market. The acceptable scientific reasons for eliminating a high dose from a study are provided in the guidelines [2-4, 6, 10] and have been discussed in Section 3.4. " | See response to Para 86 above. |
| 39 | | 4.1.4 Consistency (see Section 3.2)<br>They state that "Skin keratoacanthomas were only reported in male rats and not in female rats or male and female mice". As discussed earlier (Section 3.2), the agreement of findings across species and sexes can be used to increase the strength of evidence of a particular tumour finding but should not be used to reduce that strength of evidence without some knowledge of how these tumours arise and why. They do not provide a reasoning for why these tumours should be induced by glyphosate in both sexes or different species and in fact note that "No plausible underlying mechanism is currently identified for the induction of this tumour type". | Comparisons of incidences between sexes<br><br>In the latest version of the CLP Guidance (2017, P383-384) the relevant section states that "*Effects seen only in one sex in a test species may be less convincing than effects seen in both sexes, unless there is a clear patho-physiological difference consistent with the mode of action to explain the single sex response. However, there is no requirement for a mechanistic understanding of tumour induction in order to use these findings to support classification*".<br><br>The analysis conducted by RAC was completely consistent with the CLP Guidance and in this case it was reasonable to consider the findings less convincing when considering all the data in males and the complete absence of findings females (as well as in mice of either sex). |
| 40 | | 4.1.5 Historical Control Databases | This was simply a statement of fact – the HCD were limited for this tumour type. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | They note that "The incidences exceeded the available HCD range or from individual studies when available, however, noting that the HCD are very limited for the induction of skin keratoacanthomas in male rats" suggesting that the use of the historical control data in the evaluation of this tumour should be very limited.<br><br>They then go on to cite a single study as if it is definitive on the subject writing that "Skin keratoacanthoma is a benign tumour which is shown to be rather common in aged male rats (Zwicker et al., 1992)". They go on to note the timing of these tumours appearing in this paper [16] matches that seen in the positive studies in male SD rats. This study is used in a misleading manner to reduce the weight of evidence for these tumours. The study is also not an acceptable historical control group for any of the three positive studies in SD rats (years do not match, laboratory does not match, etc.) [6]. Also, the timing of tumour appearance is implied to be similar in treated groups to that in control so they must be spontaneous: this is not demonstrated by the ECHA and even if it was, this could be a promotional effect of glyphosate which would not change the timing, just the counts. The study by Zwicker et al. [16] demonstrates a clear difference in spontaneous tumour rates between male (3%) and female (0.3%) rats that is not used to argue why these tumours have not been seen in | The study referred to (Zwicker et al, 1992) is not cited as historical control data for the tumour type, as claimed (the actual incidences in the study were not referred to either in the opinion or in the CLH report). The point is made that the tumour type is rather common in aged male rats. The article itself notes that (in terms of skin neoplasms in aged SD rats, the topic of the article) "*Keratoacanthoma was the most frequent epithelial neoplasm in males*". This is an additional factor appropriately taken into account in the assessment and consideration of the timing was part of assessing this consideration. The differences in the incidences in the studies assessed between male and female rats reflect the differences seen in the article by Zwicker et al (1992). |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | females (too few to be promoted out) and suggests a bias in their presentation of these data. Finally, at 3% spontaneous induction in male rats, these are not "common tumours". This bias in reporting extends to the skin basal cell tumour findings since Zwicker et al (1992) report a spontaneous tumour rate of 1/717 or 0.14% for basal cell tumours which was conveniently not noted when discounting the 50-fold (4/78 or 5.1%) increase in basal cell tumours in CA 515/004 (1997). | |
| 41 | | 4.1.6 Progression of Lesions to Malignancy<br>"ECHA notes that no malignant squamous cell carcinomas (MSCC) were reported suggesting that, by not progressing from keratoacanthoma to MSCC, these tumours are not glyphosate induced".<br>"Other literature exists that support these findings that are not mentioned here (see [11] pages 11-12) as does the increased incidence of follicular hyperkeratosis in the skin noted by [11] and cited for basal cell tumours in the ECHA opinion (page 63)." | Again, this is one factor, which RAC took into account and which was therefore addressed in the opinion.<br><br>Reference [11] is to Portier, 2020. The references on pages 11-12 in that paper are to studies using glyphosate based herbicide (which, as noted in the opinion, are of limited use for investigating the carcinogenic effect of glyphosate) and in vitro studies looking at oxidative stress with human skin cells. |
| 42 | 4.2 kidney tumours | P11, last paragraph "ECHA in its Opinion (ECHA 2022, p.67) acknowledged "three positive studies" in CD-1 mice with regard to kidney tumours, but dismissed the carcinogenic effects seen in all three studies". | A few paragraphs earlier page 67 of the opinion notes as follows: "*Low, but elevated incidences of renal tumours were reported at the high dose exposures in three of the five mouse carcinogenicity studies*". This is what is meant by the "positive studies". The text in the opinion continues: "*The increases in renal tumours were not statistically significant in pairwise comparisons (Fisher's exact test), but when the Cochran-Armitage trend test was used, statistical significance was reported in these studies*." |
| 43 | | 4.2.1 Historical Control Database<br>"ECHA argued that, except for the 1997 study [17], | Apparently relevant kidney tumour data were available from memoranda from US EPA archives. The information was not directly submitted during the CLH process, |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | HCD were not supporting the study finding of an increase in tumour incidence. Overall, the opposite is true. For the 1983 study [18], ECHA (2022, p. 67), ECHA claimed that HCD were no longer available. This is not true (see "Summary of Evidence", below). In fact, the HCD for this study are available and support the study finding". | but there were references to these memoranda in Portier (2020), which was included in the evaluation conducted by the DS as well in the RAC opinion. These indicate a HCD range up to 3.3% from Bio/Dynamics studies. On this basis the findings from this study appear to have exceeded the HCD. This information has no impact on the conclusion since HCD were not a factor in the assessment of this study. An important consideration for the conclusion was the very high top dose used in this study. In relation to this, the RAC opinion stated (on P67) as follows: "*In two of the three positive studies (CA 5.5/018-019, 1997; CA 5.5/023, 1983), increased tumour incidences were only observed at very high doses (> 4000 mg/kg bw/d) at which the body weight gain in males were decreased compared to controls by up to 11% and 15% in the CA 5.5/023 (1983) and the CA 5.5/018-019 (1997) study, respectively. The OECD TG 452 for carcinogenicity studies does not give a precise top dose recommendation, but states that the highest dose level should normally be chosen to identify the principal target organs and toxic effects while avoiding suffering, severe toxicity, morbidity, or death, and the highest dose level should be chosen to elicit evidence of toxicity, as evidenced by, for example, depression of body weight gain (approximately 10%). RAC therefore gives less weight to the findings at these very high dose levels. RAC also notes that the mouse is exposed to glyphosate via the diet with a high exposure to the gastrointestinal tract. The human relevance of the renal tumours at very high doses is considered to be low and the overall evidence for the increase in renal tumours having been caused by glyphosate is considered insufficient for classification as was also concluded in the RAC opinion from 2017*." |
| 44 | | "In fact, the HCD for this study are available and support the study finding. In addition, for the 2001 study [19], more details are needed to objectively assess whether the HCD support the study results (see "Summary of Evidence", below). Even if the upper | The Regulators are required to conduct an objective assessment, which involves the application of expert judgement and weight of evidence.<br>Relevant to this is Annex I, paragraph 1.1.1.2 of CLP, which states (in part) that … "Expert judgement may also be required in interpreting data for hazard classification of substances, especially where weight of evidence determinations are needed". |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | HCD-range proves to be valid". P12 | This is further explained in Annex I, paragraph 1.1.1.3 of CLP, as follows: "A weight of evidence determination means that all available information bearing on the determination of hazard is considered together, such as the results of suitable in vitro tests, relevant animal data, information from the application of the category approach (grouping, read-across), (Q)SAR results, human experience such as occupational data and data from accident databases, epidemiological and clinical studies and well-documented case reports and observations. The quality and consistency of the data shall be given appropriate weight. Information on substances or mixtures related to the substance or mixture being classified shall be considered as appropriate, as well as site of action and mechanism or mode of action study results. Both positive and negative results shall be assembled together in a single weight of evidence determination". |
| 45 | | "OECD Guidance Document 116 [6] (section 4.22) provides guidance on using several formal statistical tests to include historical control data into the evaluation of animal carcinogenicity studies. Portier [11] provided p-values from a formal evaluation using historical controls for the 1983 [18] study (p=0.008) and the 1997 [17] study (p=0.009) that were ignored by ECHA". | As the publication referred to, Portier (2020), was extensively referred to in the CLH report (more than 50 times) as well as in the opinion (30 times), this indicates that the publication was considered in detail and therefore the fact that all figures from the document were not included in the regulatory documents does not mean that they were ignored. |
| 46 | | "RAC should have noted that there were two animals with adenomas among the 50 males of the high dose group whereas no such tumours were observed in the any other group." | The phrase is taken out of context. The statement in the opinion was referring to the statistical significance observed despite a very low incidence in the findings. The statement in full is as follows: "*RAC notes that although the p-value determined in the trend test in the study CA 5.5/018-019 (1997) indicated that the finding was statistically significant, there were only two adenomas among the 200 males examined in this study*" (page 67 of the opinion). |
| 47 | | 4.2.2 "In addition, progression to malignancy, i.e. the conversion from a benign to a malignant | The authors avoid the fact that the 1993 study, in which there were renal tumours in 2 control animals as well as in 2 low dose animals, but none at the two higher |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | tumour, is time-dependent. In other words, a longer study duration increases the likelihood of such a progression. ECHA failed to take into consideration that the two studies with "no evidence" (studies of 1997 [17] and 2001 [19]) were only of 18 months duration whereas the study with "equivocal" data [18] was of 24 months duration." | doses, was also a 24 month study. This reinforces the likelihood that the tumours observed were attributable to chance, because while the higher incidence in some studies was at the higher doses, in others (such as this one) the highest incidences were in the lowest dose and the control groups. |
| 48 | | 4.2.3 limit dose | Issues raised are addressed above (see the response to Paragraph 86). |
| 49 | | 4.2.4 Mode of action<br>"Lack of knowledge about mode of action should not be used to weaken the evidence as ECHA [8] (p.53) did by claiming that "there was no plausible mechanism" to explain how the kidney tumours were elicited. In addition, this is not true. First of all, there is evidence for genotoxicity in somatic cells which was largely dismissed by ECHA. Moreover, oxidative stress is known as one of ten key characteristics of carcinogens [20], and was also acknowledged several times in ECHA's Opinion [8]".<br><br>And<br><br>"While studies on oxidative stress were acknowledged in general, neither oxidative stress in general nor the Gao et al. [22] study in particular were taken into consideration in the Section on "Mechanistic studies from the public literature" [8] (p.54/55). Instead ECHA incorrectly claimed a lack of a plausible mechanism for kidney tumours. | The main claim in this section is that oxidative stress was not adequately taken into account during the assessment of RAC. In the context of the CLP criteria the primary source of evidence to inform on classification is enumeration of tumours in animal studies and determination of their level of statistical significance. Many other factors can be taken into consideration including mode of action/mechanistic considerations.<br>Oxidative stress is a mechanism that can lead to tumour formation and therefore falls into the latter category as a factor that can be taken into consideration when assessing tumour incidences.<br>ECHA's independent assessment is based on a large number of scientific studies designed to examine the hazardous properties of glyphosate, including whether it causes cancer. All available evidence was carefully examined to arrive at a conclusion. No important findings were dismissed. Tumour incidences in the available studies were examined in detail and the conclusion was that there was no convincing evidence that glyphosate induces tumours.<br>In the absence of clear evidence of tumours linked to glyphosate, evidence that glyphosate causes oxidative stress is not relevant for the conclusion. Findings of oxidative stress in a study are not on their own sufficient for classification. In particular, potential mode of action considerations arising from one study cannot provide support in the absence of convincing evidence for carcinogenicity in another study. The mechanistic data from the Gao study were included in the CLH report and considered in the RAC opinion. Furthermore, the arguments put forward |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | | by the authors of the Annex were heard and considered by RAC during the opinion development process. |
| 50 | | 4.2.5 Consistency<br>"Portier [11] tested for a common trend in kidney adenomas, carcinomas and combined adenomas and carcinomas using pooled data in CD-1 mice (including one study excluded by ECHA [25]) and saw p-values of 0.006, 0.031, <0.001 respectively. This was not mentioned by ECHA [8]." | As the publication referred to, Portier (2020), was extensively used in the CLH report (more than 50 times) as well as in the opinion (30 times), this indicates that the publication was considered in detail and therefore the fact that all figures from the document were not included in the regulatory documents does not mean that they were ignored. |
| | | 4.3 Malignant lymphoma | |
| 51 | | 4.3.1 Strength of evidence<br>"As demonstrated in the table provided on page 68/69 in [8], three studies exhibited significantly increased tumour incidences when the assessment was made with proper statistical methods. This applies to the use of the Cochran Armitage trend test for [17] and [24], and the Peto-test for [19] because of slight mortality differences between groups. ECHA [8] (p.70) then states that no statistical significance was observed in any of the studies using Fisher's exact test as a 2-sided test (see Section 2.0). This is an attempt to reduce the importance of the trend tests disregarding OECD guidance document 116 [6] (p. 123)." | This is a statement of fact concerning the statistical analyses, which is not disputed by the authors. |

**Relevant scientific arguments provided in the review letter submitted by Aurelia Stiftung for the active substance glyphosate and the conclusions drawn by ECHA on the specific points raised**

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| 1 | **11. Incorrect assessment of the risk of cancer (P40)** | **(1)** "In the view of the applicants, the risk assessment is also flawed because EFSA wrongly bases its conclusions on ECHA's inclusion of glyphosate as 'not carcinogenic'. ECHA has overlooked important evidence on carcinogenicity and neglected evidence that glyphosate is causing oxidative stress".<br><br>Annex 27 Clausing et al., Glyphosate and Oxidative Stress: ECHA's superficial approach neglects existing hazards, 2023<br><br>**(2)** "In addition, numerous other shortcomings in the assessment of carcinogenicity by ECHA and EFSA are to be criticised. In its opinion of 30 May 2022, ECHA, among other things, rejects study results due to an alleged 'limit dose' of 1 000 mg/kg. However, test Guideline 451 and OECD Guidance 116 do not include this 'limiting dose'".<br><br>**(3)** Annex 29 Pan Europe, EU glyphosate | ECHA's responses to the central issues raised in these documents (on the role of oxidative stress and the dose of 1000 mg/kg) have been published on the ECHA website since October 2023 (7e7b03bc-b2f1-8949-b6f6-da9feb1f7292 (europa.eu)). The responses are summarized below:<br><br>**(1) "Oxidative stress"**<br>The main claim on this point is that "oxidative stress was not adequately taken into account during the assessment of ECHA's RAC, leading to underestimation of the potential of glyphosate to cause cancer". Firstly, it is useful to explain that in the context of the CLP criteria the primary source of evidence to inform on classification is enumeration of tumours in animal studies and determination of their level of statistical significance. Many other factors can be taken into consideration including mode of action/mechanistic considerations. Oxidative stress is a mechanism that can lead to tumour formation and therefore falls into the latter category as a factor that can be taken into consideration when assessing tumour incidences. ECHA's independent assessment is based on a large number of scientific studies designed to examine the hazardous properties of glyphosate, including whether it causes cancer. All available evidence was carefully examined to arrive at a conclusion. No important findings were dismissed. Tumour incidences in the available studies were examined in detail and the conclusion was that there was no convincing evidence that glyphosate induces tumours.<br><br>In the absence of clear evidence of tumours linked to glyphosate, evidence that glyphosate causes oxidative stress is not relevant for the conclusion. Findings of oxidative stress in a study are not on their own sufficient for classification. In particular, potential mode of action considerations arising from one study cannot provide support in the absence of convincing evidence for carcinogenicity in another study. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | evaluation fails to accept key mechanism that can lead to cancer, 2023. This Annex included references to a letter to Commissioner Stella Kyriakides dated 7 September 2023 with the heading "Subject: Stop the reapproval of glyphosate due to major deficiencies in carcinogenicity assessment". In addition to also raising the issues relating to "oxidative stress" and "limit dose", additional points made in the letter the following headings: "Missing industry genotoxicity studies", "Tumour incidences were observed in glyphosate cancer studies" and "Malignant lymphomas in animal studies complement the evidence in epidemiology studies".<br><br>**(4)** Annex A.28 Presentation by Dr Peter Clausing, Pesticide Action Network (PAN) Germany, 2023 | The mechanistic data from the Gao study were included in the CLH report and considered in the RAC opinion. The arguments put forward by the authors of the publication (Clausing et al, 2023) were heard and considered by RAC during opinion making.<br><br>**(2) "Limit dose"**<br><br>The deliberations of RAC on this particular issue are clearly and transparently set out in the published opinion, therefore, there is no deception. RAC did not dismiss the tumour findings at doses above 1000 mg/kg bw/day, but the findings at the very high doses (above 4000 mg/kg bw/d) were given lower weight, for the reasons explained in the Opinion as well as in the CLH report. In short, while there were low incidences of tumours at the highest doses in these studies, this was in combination with other effects (body weight gain data suggesting general toxicity) and therefore RAC, following recommendations in the relevant OECD guideline, gave these findings lower weight amongst all the other information available to inform on carcinogenicity.<br><br>**(3) Other issues raised "Missing industry genotoxicity studies"**<br>As regards the concern that key tests were not conducted due to a statement in the RAC opinion referring to the absence of specific assays in relevant target organs (OECD TG 489 "the comet assay" and OECD TG 488 "TGR"), firstly, it should be noted that the CLH process assesses available data – there is no mechanism to generate additional information. Secondly, please note that ECHA has addressed these particular issues in a letter to Bas Eickhout MEP, who raised this in the Exchange of views on 11 July 2022. ECHA addressed these concerns in our letter as follows (available at Letter):<br>"*The statement quoted from the opinion related to the Comet assay and Transgenic rodent (TGR) somatic and germ cell gene mutation assays which are two particular assays among many other lines of evidence potentially informing a classification. The opinion noted the absence of these assays/studies in relevant tissues, but also noted that the biological importance of such DNA lesions (i.e., as identified from these assays) in relation to mutagenicity is* |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | | *equivocal, therefore the fact that some studies of this type were not included is not crucial for the conclusion*"<br>And<br>"*the data available for evaluation of germ cell mutagenicity is extensive and includes studies covering bacterial and mammalian cell in vitro mutagenicity assays as well as in vivo mammalian mutagenicity assays and even some human data. Furthermore, according to the opinion, the data includes studies of sufficient reliability and relevance to allow a robust evaluation, especially in the perspective of the requirements of the CLP Regulation. In RAC's view, the data were sufficient to arrive at a robust conclusion without these assays/studies*."<br><br>**"Tumour incidences were observed in glyphosate cancer studies"**<br>The reference for this issue as cited in the letter is the publication by HEAL on 8 June 2022.<br>There is a detailed response to the claims made in this publication on ECHA's website (at Response).<br><br>While we welcome the opportunity to further increase transparency about the reasoning in the RAC opinion on certain issues, we do not agree with the conclusions of the HEAL report and consider the criticisms unfounded for the reasons explained in our published response.<br><br>RAC experts, in accordance with their mandate, applied the CLP Regulation's criteria to toxicological and epidemiological findings and weighed all the evidence in arriving at their conclusions on classification. They considered the strength of the statistical evidence, dose-response relationships, concurrent and historical control data and the biological relevance of the findings.<br><br>**"Malignant lymphomas in animal studies complement the evidence in epidemiology**<br>**Studies"**<br><br>RAC concluded that the lymphoma incidences in male mice showed a slight, but clearly variable increase, but the biological and human relevance of the |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | | findings is uncertain because (among other reasons)<br>• The maximum incidences were mostly within the available historical control range<br>• The increases tended to be confined to the highest dose<br>• The increases were not seen in female mice (or in rats)<br><br>Attempts have been made to draw a link between the findings in mice to non-Hodgkin's lymphoma in humans.<br>The role of non-Hodgkin's lymphoma was addressed in detail in the RAC opinion. No association between exposure to glyphosate-based herbicide and non-Hodgkin's Lymphoma was found in the AHS cohort study, which is the only prospective cohort study available (ref Andreotti et al, 20181) and was considered by RAC as the most robust epidemiological study since it includes appropriate controls, a balanced assessment, and due consideration of bias or confounding factors. Weak positive associations have been observed in some case-control studies (but not consistently) and in meta-analyses (which depend on assumptions made about both exposure level and latency period).<br><br>RAC agreed with the dossier submitter that there is no epidemiological evidence of an association between exposure to glyphosate-based herbicide and the risk of Hodgkin's Lymphoma.<br><br>Considering the lack of evidence for biological and human relevance of the findings of malignant lymphoma following exposure to glyphosate in animal studies and the absence of epidemiological evidence of an association between exposure to glyphosate-based herbicide and the risk of Hodgkin's Lymphoma, the argument that there is a link between the findings in animals and humans is untenable.<br><br>**(4) Weight of evidence**<br><br>The points raised in the presentation from PAN Germany (Annex A.28) "Glyphosate and ECHA's "weight of evidence"" have been addressed by ECHA in their document "Relevant scientific arguments provided in the review letter submitted by PAN & others for the active substance glyphosate and the |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>ECHA's scientific views on the specific point |
|---|---|---|---|
| | | | conclusions drawn by ECHA on the specific points raised". A detailed response to the claims made are also available from ECHA response the "HEAL report" publication on ECHA's website (at Response). |

## Relevant scientific arguments provided in the review letter submitted by PAN & others for the active substance glyphosate and the conclusions drawn by EFSA on the specific points raised

*It should be noted that the original request for internal review was provided in French language. Where available, a complimentary English translation has been provided to EFSA and ECHA by the Commission for the purpose to facilitate assessment by the Agencies. The English translation as displayed in column 2 has been generated by using an automated machine translation tool. Therefore the quality and accuracy of the translation may vary from the original text and should not be regarded as official translation. Only the original text of the request submitted in French should be considered as the authentic text.*

*For the footnotes mentioned in the table (column 2), refer to the original request for internal review 'Demande de réexamen interne du Règlement d'exécution 2023/2660 de la Commission du 28 novembre 2023 renouvelant l'approbation de la substance active glyphosate' (page 51 onwards).*

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| 1. | Paragraphs 31-34<br><br>Illegalities and manifest errors in the assessment of the genotoxicity of glyphosate<br><br>Serious indications of genotoxic potential from independent scientific literature systematically questioned or ignored | # 34. The vast majority of independent peer-reviewed scientific studies on the subject conclude that glyphosate has a genotoxic potential (see table in **Annex 1**). In 2015, the International Centre for Research on Cancer ('IARC') concluded that there was '*strong evidence for genotoxicity'* on the basis of an in-depth review of 118 independent studies, 70 % of which were positive[50]. In 2021, INSERM considered that "*while the results obtained with genotoxicity and mutagenicity tests are taken into account as a whole, many studies were published with rather positive results on a genotoxic effect*"[51]. In its contribution to the public consultation following the EFSA conclusions, INSERM stated that '*the studies showing that glyphosate has genotoxic effects are more important in terms of quality and quantity than those suggesting an absence of effect. A genotoxic effect of glyphosate is* | The weight of evidence approach for genotoxicity on glyphosate during the ECHA RAC assessment and the peer review included more than 70 studies (regulatory and public literature studies) assessed as acceptable, supplementary or supportive (see Renewal Assessment Report at Table 2.0.5.2-3 and Table 2.0.5.2-4 (RAR, Volume 1, 2023)[8]).<br><br>The position of the French National Institute of Health and Medical Research (Inserm), 2021 was considered during the recent peer review process (see data requirement 2(51) in Part 2_Peer Review Report_Glyphosate_reporting tables_public[9]). The conclusion of the peer review is reported below in the reply to paragraphs 37-38. |

[8] available in the Open EFSA, 'Supporting documents' section under EFSA Question number EFSA-Q-2020-00140: https://open.efsa.europa.eu/study-inventory/EFSA-Q-2020-00140 (refer to Glyphosate_Final RAR_public.zip)

[9] Available in the Peer review Report in Open EFSA, Supporting documents section under EFSA-Q-2020-00140 (Part 2_Peer Review Report_Glyphosate_reporting tables_public, electronic page 2417 of 2930)

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | INSERM | *consistent with the induction of oxidative stress, observed in different species and cell systems, sometimes at exposure doses consistent with those encountered in the environment*"[52]. | |
| 2. | Paragraphs 35-36<br><br>Illegalities and manifest errors in the assessment of the genotoxicity of glyphosate<br><br>Serious indications of genotoxic potential from independent scientific literature systematically questioned or ignored | # 35. It is apparent from volume 3 (B.6.4) of the RAR that only around fifteen of these studies were listed by applicants for re-approval without the rapporteur Member States having considered it necessary to request the submission of other studies, including some which, although prior to a ten-year period[53], continue to be relevant. This is particularly problematic if it is to be remembered that, at the time of the previous renewal (2017), these older studies had <u>not</u> been examined by the RMS. Indeed, it appeared at that time that, instead of carrying out an independent review of the scientific literature, the RMS had merely reproduced verbatim the presentation, interpretation and evaluation made of it by the applicants for re-approval. As Dr Weber notes (**Annex 2**), " *in the incriminating passages the reader has no doubt that the BfR is describing its own literature research – including presenting its methodology – and giving its own judgments, while in reality these are the judgments either of the "Glyphosate Task Force" or of Dr Kier*". <u>This means that the majority of studies published more than a decade ago and which served as the basis for the IARC conclusions referred to above were never reviewed by the competent authorities, neither during the current nor during the previous renewal process.</u> | During the peer review EFSA asked further clarifications to both the applicant and the RMS on the literature search conducted (see EFSA conclusion on data requirements (general) 2.62 and 2.63 and related action points for the RMS in Part 4_Peer Review Report_evaluation tables_July 2023 [10] ). These included further clarifications on the approach used for the relevance and reliability assessment of published and guideline studies, and further consideration of studies conducted outside the timeframe covered by the literature search as well studies previously included in the former RAR or in the addenda of the RAR (Germany, 2015). The addenda of the RAR (Germany, 2015) included the German assessment of the monograph published by the International Agency for Research on Cancer (IARC). |

[10] Available in the Peer review Report in Open EFSA, Supporting documents section under EFSA-Q-2020-00140 (Part 4_Peer Review Report_Glyphosate_evaluation tables_public, refer to section 2, electronic page 282-288 of 1093).

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | # 36. Moreover, none of the studies considered under the independent literature were found to be "*relevant and reliable*" or "*relevant and reliable with restrictions*", so none was used as direct support for risk assessment and hazard identification. At best, some studies were considered "*additional*", i.e. "*less relevant and reliable with restrictions*". The same fate was given to the numerous studies sent to the competent authorities in the context of the public consultation. For many studies, the ranking initially proposed by the applicants was even revised downwards by the RMS. The reasons given for sub-weighting or even excluding these studies are mainly due to *(i)* the fact that they implement test systems not standardised by these *Test Guidelines* – TG (e.g. insects, worms, amoebas, fish or plants), or *(ii)* that they relate to glyphosate-based formulations and not to glyphosate itself. | |
| 3. | Paragraphs 37-38<br><br>Illegalities and manifest errors in the assessment of the genotoxicity of glyphosate<br><br>Serious indications of genotoxic potential from independent scientific literature | # 37. As INSERM observes, it is therefore the failure to take into account results from non-standardised test models and tests carried out with formulations that alone explains EFSA's conclusions on the absence of genotoxicity:<br>The difference in opinion between the Inserm collective expert review and the RAR on the question of genotoxicity stems from the fact that the Inserm collective review takes into account both the results using non-standard models (i.e., non-mammalian models such as fish and crustaceans; not considered for classification in the RAR), and those obtained | The position of the French National Institute of Health and Medical Research (Inserm), 2021 was considered during the peer review process (see data requirement 2(51) in Part 2_Peer Review Report_Glyphosate_reporting tables_public).<br><br>The assessment of genotoxicity is based on a weight of evidence (WoE) approach (EFSA Scientific Committee, 2011[11]).<br><br>The WoE approach for genotoxicity on glyphosate active substance during the peer review included more than 70 studies (regulatory and public literature studies) assessed as acceptable, supplementary or supportive (see Renewal |

[11] EFSA Scientific Committee, 2011. Scientific Opinion on genotoxicity testing strategies applicable to food and feed safety assessment. EFSA Journal 2011;9(9):2379, 69 pp. https://doi.org/10.2903/j.efsa.2011.2379

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | systematically questioned or ignored<br><br>INSERM | with formulations (GBHs) that better reflect the reality of exposure in humans[54].<br><br># 38. It should be noted, however, that *(i)* the studies provided by applicants for re-approval are also not in line with the TG in force (see point 44 *below*); *(ii)* in any event, TGs, which are adopted in consultation with industry, do not favour the 'sensitivity' of studies and do not reflect the latest scientific knowledge[55]; *(iii)* the formulation studies are relevant, on the one hand because they can provide decisive insights into the toxicity of the representative formulation and, on the other hand, because, as INSERM points out, they provide information on the effects of exposure to glyphosate in real life. As noted by the European Parliament's Legal Service, '*it is clear that active substances need to be assessed not in abstract terms, as isolated substances, but in their quality of future PPP contents*'[56]. It is remarkable and worrying that the competent authorities did not even consider it necessary to examine the composition of the tested formulations before rejecting the studies concerned as irrelevant. | Assessment Report at Table 2.0.5.2-3 and Table 2.0.5.2-4 (RAR, Volume 1, 2023)).<br><br>In the WoE, higher relevance was given to mammalian species compared to non-mammalian species such as fish, as well as higher relevance was given to those studies validated or performed according to agreed international protocols, not the case for fish genotoxicity studies (see point 2(44) in Part 2_Peer Review Report_Glyphosate_reporting tables_public[12]).<br><br>As regards the composition of the tested formulations: where available, the applicants provided information on the composition of the formulations (different from the representative one) used in published and non-published studies. Considerations on whether these formulations were comparable to the formulation for the representative uses were also included in the RAR Volume 4. Depending on the availability of the evidence for the different toxicological endpoints, e.g. DNT, studies conducted with different salt-forms and/or formulations other than the representative one, were considered for their reliability and relevance and discussed as part of the WoE in the risk assessment (see Data requirement (general) 2.62 in Part 4_Peer review report_evaluation table (section 2)). For genotoxicity, the contribution of studies performed with formulations containing glyphosate, including the formulation for representative uses, to the WoE for genotoxicity of glyphosate was as follows: glyphosate is the active substance in many formulations. Formulations may inform on the genotoxicity of glyphosate and therefore studies with the formulation should be considered for the |

[12] Available in the Peer Review Report in Open EFSA, Supporting documents section under EFSA-Q-2020-00140 (Part 2_Peer Review Report_Glyphosate_reporting tables_public, electronic page 2383 of 2930).

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | | assessment of glyphosate. However, lower weight was given to studies with formulation to assess the genotoxicity of glyphosate, given the high uncertainties regarding potential different components of the formulation, not only glyphosate (Peer Review Report_Annex 3 of the Pesticides Peer Review Experts' Meeting TC 80[13]).<br><br>The formulation for the representative uses is unlikely to be genotoxic or mutagenic (based on a WoE approach and considering the study performed with the formulation for the representative uses (EFSA, 2023)[14]. |
| 4. | Paragraphs 39-41 Illegalities and manifest errors in the assessment of the genotoxicity of glyphosate | *Paragraphs linked to classification criteria and elements falling in the remit of ECHA*.<br><br>See ECHA response in PAN comment No 1 ("*Paragraphs 39-41 (as relevant)*") in Appendix A above. | EFSA agrees with the conclusions reached in the ECHA RAC Opinion. The outcome of the assessment of the genotoxic potential reached in the EFSA peer review based on a weight of evidence approach is in line with ECHA RAC assessment. |
| 5. | Paragraphs 42-44 Deficient regulatory studies | # 43. This methodological bias consisting, on the one hand, of excluding or sub-weighting a large number of studies conducted in independent, peer-reviewed academic institutes published in peer reviewed journals and, on the other hand, of giving the greatest credit to unpublished, commissioned and submitted tests by producers of glyphosate and glyphosate products, is manifestly contrary to the principles of completeness, excellence and – above all – independence which are supposed to guide the assessment of the harmfulness of a substance. | In the area of pesticides, the body of evidence that EFSA and the Rapporteur Member States have at their disposal is always composed of both regulatory studies, conducted according to internationally agreed OECD Test Guidelines and studies from publicly accessible literature, as required by the applicable legislation.<br><br>All such studies are assessed by the RMS, and then by the peer review, according to their scientific relevance and reliability, and this is done in line with the EFSA Guidance |

[13] Available in the Peer review Report in Open EFSA, Supporting documents section under EFSA-Q-2020-00140 (Part 3_Peer Review Report_Glyphosate_Annexes: Peer Review Report_Glyphosate_Annexes_TC80_public.pdf)

[14] EFSA (European Food Safety Authority),Alvarez, F., Arena, M., Auteri, D.,Binaglia, M., Castoldi, A. F., Chiusolo, A., Crivellente, F., Egsmose, M., Fait, G., Ferilli, F., Gouliarmou, V.,Nogareda, L. H., Ippolito, A., Istace, F., Jarrah, S., Kardassi, D., Kienzler, A., Lanzoni, A.,...Villamar-Bouza, L. (2023). Peer review of the pesticide risk assessment of the active substance glyphosate. EFSA Journal, 21(7),1–52, https://doi.org/10.2903/j.efsa.2023.8164

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | # 44. By way of illustration of this methodological bias, it should be noted, for example, that studies provided by applicants for re-approval were considered relevant, acceptable and reliable even though they had major deviations from the TG (incomplete historical control data, errors in the number of cells evaluated, failure to mention the weight of the animals tested, etc.).<br><br>CA 5.4.1/027 In vitro CA test *(acceptable: reliable with restriction/relevant):* see details on the deviations provided in the review request.<br><br>CA 5.4.2/009 In vivo MN test *(acceptable: reliable with restriction/relevant):* see details on the deviations provided in the review request. | (EFSA, 2011 and its Appendix published in 2019[15], and EFSA Scientific Committee 2017[16]) for all the active substances evaluated by EFSA.<br><br>EFSA notes that, according to EFSA Scientific Committee, 2017, *"the reliability is defined as the extent to which the information comprising a piece or line of evidence is correct. The reliability of a study may be assessed by considering the uncertainty of the evidence and everything that contributes to that uncertainty should be included when assessing reliability"*. Therefore, in line with this definition, deviation from the OECD TG is taken into account in the overall assessment for reliability to conclude on whether the latter may be impacted or not. |
| 6. | Paragraph 45<br><br>Deficient regulatory studies in contrast with studies from published literature<br><br>Predominant weight to industry studies | # 45. The contrast is striking with studies from published literature which were found to be '*less relevant*' and '*additional*':<br><br>**De Almeida et al., 2018: in vitro Comet assay (supplemental: less relevant and reliable with restrictions):** *see further details provided in the review request.*<br><br>**Mladinic et al., 2009b In vitro MN test (supplemental: less relevant and reliable with restrictions):** *see further details provided in the review request.* | The weight of evidence approach for genotoxicity on glyphosate during the peer review included more than 70 studies (regulatory and public literature studies) assessed as acceptable, supplementary or supportive (see Renewal Assessment Report at Table 2.0.5.2-3 and Table 2.0.5.2-4 (RAR, Volume 1, 2023)).<br><br>Please also refer to the other replies above and below. |

[15] EFSA (European Food Safety Authority), 2011. Submission of scientific peer-reviewed open literature for the approval of pesticide active substances under Regulation (EC) No 1107/2009. EFSA Journal 2011;9(2):2092, 49 pp. doi:10.2903/j.efsa.2011.2092

[16] EFSA Scientific Committee, 2017. Scientific Opinion on the guidance on the use of the weight of evidence approach in scientific assessments. EFSA Journal 2017;15(8):4971, 69 pp. https://doi.org/10.2903/j.efsa.2017.4971

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | **Mladinic et al., 2009 in vitro comet and MN test (supplemental: less relevant and reliable with restrictions):** *see further details provided in the review request.* | |
| 7. | Paragraph 46<br><br>Predominant weight to industry studies | # 46. Two of the three studies mentioned above are considered less relevant because they use a type of test that is not subject to TG. It is not apparent either from EFSA's guidance document on the submission of independent literature[71] or from its guidance document on *weight of evidence*[72] that that criterion should play any role in the assessment of relevance. On the contrary, the obligation to take account of independent literature is precisely intended to also examine the results of tests different from regulatory tests. Furthermore, it is not clear how a TG deviation test would be more relevant than a test carried out in the absence of TG but according to a scientifically accepted and peer-reviewed methodology. Moreover, the assertion that the *in vitro* comet test is not considered (by whom?) as a standard test method for pesticide active substances is in no way substantiated. | Genotoxicity studies on glyphosate were assessed by considering the European Union (EU) legal data requirements for pesticide active substances (Regulation (EU) No 283/2013) and following the recommendation of EFSA scientific documents on genotoxicity (EFSA Scientific Committee 2011[17], 2017[18], 2021[19]).<br><br>According to the EU data requirements, the data package should address the three apical genotoxicity endpoints: gene mutation, clastogenicity and aneugenicity. DNA damage, as measured by Comet assay, is not an apical endpoint as DNA damage can be repaired. Although the in vivo Comet assay can be a suitable follow up for substances positive in vitro in the gene mutation or clastogenicity assays (EFSA Scientific Committee, 2011), the nature of the endpoint, DNA damage, is also considered in the overall weight of evidence approach.<br><br>The weight of evidence assessment for genotoxicity on glyphosate during the peer review included more than 70 studies (regulatory and public literature studies) assessed |

[17] EFSA Scientific Committee, 2011. Scientific Opinion on genotoxicity testing strategies applicable to food and feed safety assessment. EFSA Journal 2011;9(9):2379, 69 pp. https://doi.org/10.2903/j.efsa.2011.2379.

[18] EFSA Scientific Committee, 2017. Scientific Opinion on the clarification of some aspects related to genotoxicity assessment. EFSA Journal 2017;15(12):5113, 25 pp. https://doi.org/10.2903/j.efsa.2017.5113.

[19] EFSA Scientific Committee, 2021. Guidance on aneugenicity assessment. EFSA Journal 2021;19(8):6770, 27 pp. https://doi.org/10.2903/j.efsa.2021.6770.

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | | as acceptable, supplementary or supportive (see Renewal Assessment Report at Table 2.0.5.2-3 and Table 2.0.5.2-4 (RAR, Volume 1, 2023)). |
| 8. | Paragraph 49 | # 49. …the RMS and EFSA should have examined in particular detail all the data submitted by the applicants for re-approval. However, this was not the case. No additional studies were requested from the applicants or commissioned by the European authorities. | A robust assessment of all available data has been undertaken in the context of the EU peer review, which is an iterative process starting with verification of the submitted dossier by the RMS, who may ask for further information. EFSA and the Member States are subsequently in charge of the peer-review process. This included also a rigorous evaluation of both industry studies submitted by the applicants and studies found in public literature, which were equally assessed for their relevance and reliability for the risk assessment and were taken into account in a weight of evidence approach**.**<br><br>To allow a transparent assessment of all the submitted studies, including the regulatory studies from the applicants, the RMS was asked to transparently report both the assessment of the reliability of the studies and the relevance of the study results to conclude on the overall weight of evidence.<br><br>Following the categorisation of toxicology standard studies as acceptable, supplementary supportive and not acceptable (see RAR, volume 1, level 2), the weight of evidence approach for genotoxicity on glyphosate during the peer review included more than 70 studies (regulatory and public literature studies) assessed as acceptable, supplementary or supportive (see Renewal Assessment Report at Table 2.0.5.2-3 and Table 2.0.5.2-4 (RAR, Volume 1, 2023)).<br><br>Overall, all available studies, both from the applicants and from publications, have been duly considered and assessed for their relevance and reliability following a rigorous |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | | approach as detailed in the RAR. Where needed, additional information has been requested during the regulatory stop of the clock to complete the data package and address the comments received during the public and targeted MS consultation.<br><br>With regard to the genotoxicity assessment, mentioned in this and the preceding paragraphs, EFSA did identify several data requirements for the applicants, please refer to the Reporting table (section 2) points 2(149), 2(151), 2(152), 2(153), 2(154), 2(155), 2(158), 2(161), 2(165), 2(169), 2(201), 2(202), 2(203), 2(204) and Reporting table identified following public comments (section 2) points 2(43), 2(44), 2(51).<br><br>In addition, a dedicated expert consultation on all relevant endpoints took place, including specific discussion on the impact of the identified deviations of the genotoxicity studies on glyphosate (considerations of major vs minor deviations). The experts' consultation resulted in the collegial agreement by the experts on the relevance and reliability of the studies, taking into account deviations from the applicable test guidelines (TGs).<br><br>Overall, the genotoxicity data package on glyphosate was considered extensive and sufficiently robust that permit proper conclusions to be drawn on genotoxicity without the need for requesting further vertebrate studies**.** |
| 9. | Paragraphs 50-51<br><br>Studies provided by the applicants are affected by numerous flaws | # 51. On the one hand, an independent evaluation (**Annex 10**) shows that, out of the 24 studies provided by the re-approval applicants and considered reliable or reliable with restrictions and used by the competent authorities during the previous re-approval procedure, only <u>two</u> studies provided by the applicants should be classified as reliable without restrictions (**Annex 3)** | Studies were scored for their relevance and reliability, resulting in studies being either acceptable, supportive, supplemental or not acceptable. The studies that were considered not acceptable are not taken into account for the overall weight of evidence assessment on genotoxicity of glyphosate. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | while the other studies should have been considered partially reliable only. | The weight of evidence assessment for genotoxicity on glyphosate during the peer review included more than 70 studies (regulatory and public literature studies) assessed as acceptable, supplementary or supportive (see Renewal Assessment Report at Table 2.0.5.2-3 and Table 2.0.5.2-4 (RAR, Volume 1, 2023)). |
| 10. | Paragraphs 53-54<br><br>Incomplete genotoxicity testing | # 54. The refusal by the competent authorities to request that other tissues be tested is incomprehensible, not only in the light of the current state of independent scientific research, but also in the light of **EFSA's** own positions. In its **Scientific Opinion on genotoxicity testing strategies,** EFSA states very clearly: " *The classical in vivo tests may be limited to certain tissue restrictions (bone marrow, peripheral blood cells, hepatocytes). It is obvious that the value of the in vivo tests increases if more than one tissue is tested. Therefore, tests without obvious tissue restriction should be recommended as follow-up tests, where possible*[79]. Similarly, **EFSA stresses the value of the Comet Assay test:** " *the in vivo COMET assay is considered a useful indicator test in terms of its sensitivity to substances which cause gene mutations and/or structural chromosomal aberrations and can be used with many target tissues. [...] The in vivo COMET assay has been suggested by several authors (Tice et al., 2000; Hartmann et al., 2003; Burlinson et al., 2007) as a suitable follow-up test to investigate the relevance of positive in vitro tests (gene mutagens and clastogens, but not aneugens)*[80]. However, *in vitro* tests were positive, so that it was necessary to use a Comet Assay test. This view is shared by ANSES, according to which: " *While almost all in vivo tests lead to non-statistically significant results, there is no in vivo comet test results, which seem to be the most sensitive* | The EFSA Scientific Opinion on genotoxicity testing strategies (EFSA Scientific Committee, 2011) recommends a WoE approach. The WoE for genotoxicity on glyphosate during the peer review included more than 70 studies (regulatory and public literature studies) assessed as acceptable, supplementary or supportive (see Renewal Assessment Report at Table 2.0.5.2-3 and Table 2.0.5.2-4 (RAR, Volume 1, 2023)).<br><br>The EFSA Scientific Opinion on genotoxicity testing strategies (see chapter 4.1) considered that indicator tests (e.g. Comet assay) detect pre-mutagenic lesions, which may not result in mutations, e.g. repairable DNA damage measured by the Comet assay.<br><br>This consideration was part of the discussion during the Pesticides Peer Review Experts' meeting TC 80 (see experts' consultation point 2.1 identified following comments by public):<br><br>*"In the overall WoE for genotoxicity, more weight is given to apical endpoints, gene mutation and chromosome aberrations (i.e. permanent DNA lesions), than to primary DNA damage (may be transient or reversible). This is also in line with the European Union data requirements where the data package should address the three apical genotoxicity endpoints: gene mutation, clastogenicity and aneugenicity. Primary DNA damage, as measured by Comet assay, is not an apical endpoint as DNA damage may* |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | *biological parameter. Therefore, it might be useful to perform an in vivo Comet Assay test on defined target organs (kidney and liver)"*[81]. Finally, it is pungent to note that in its opinion on the carcinogenicity of glyphosate, ECHA itself acknowledges doubts as to the genotoxic nature of glyphosate, deploring the absence of an *in vivo* comet test and the examination of tissues other than spinal cord[82]. | *be transient or reversible. Although the in vivo Comet assay can be a suitable follow up for substances positive in vitro in the gene mutation or clastogenicity assays (EFSA SC, 2011), the nature of the endpoint, primary DNA damage, is also considered in the overall WoE"*<br><br>Overall, the genotoxicity data package on glyphosate was considered extensive and sufficiently robust to reach proper conclusions on genotoxicity **without the need for requesting further vertebrate studies**. |
| 11. | Paragraphs 54, 56<br><br>Incomplete genotoxicity testing | *Paragraphs linked to classification criteria and elements falling in the remit of ECHA*.<br><br>See ECHA response in PAN comment No 2 ("*Paragraph 54 (last sentence) and paragraph 56*") in Appendix A above. | Overall, the genotoxicity data package on glyphosate was considered extensive, robust and allowing proper conclusions to be drawn on genotoxicity.<br><br>All pertinent studies were also part of the hazard assessment undertaken in the context of the formal assessment of the proposal for harmonised classification and labelling in accordance with Regulation (EC) No 1272/2008 conducted by ECHA in parallel to the EFSA peer review. When carrying out the risk assessment in the framework of the peer review, EFSA adopted ECHA's hazard assessment and the conclusions of the ECHA Committee for Risk Assessment (RAC) on harmonised classification and labelling delivered in their Opinion on 30 May 2022 (ECHA, 2022).<br><br>The weight-of-evidence assessment conducted in the EFSA peer review was in line with the ECHA RAC outcome**.** |
| 12. | Paragraphs 57-58<br><br>The genotoxic potential of an impurity | # 57. In its conclusion, EFSA considers that one of the impurities resulting from the glyphosate production process, namely glyphosine, showed a potential for clastogenicity in an in *vitro chromosomal aberration assay*[84]. It further notes that that test was not followed up by an *in vivo* test, as it should have been. EFSA infers a lack of data. | EFSA identifies a critical area of concern by considering:<br><br>1) the conclusions on the relevance of impurities and,<br>2) The proposed maximum content of the relevant impurity in the specification and, |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | # 58. This classification ("*lack of data*") reflects a manifest error of assessment. In other dossiers [*cypermethrin, isoflucypram, benfluralin, see in footnote 85*], EFSA considered the genotoxic potential of an impurity to be a 'critical area of concern', meaning that the active substance under review is unlikely to meet the criteria for the protection of human health and the environment set out in the PPP Regulation[85]. In any event, this lack of data reveals a new source of uncertainty as to the genotoxicity of glyphosate. | 3) the evidence on whether the batches used in toxicity testing are representative or not of the reference specification, or,<br>4) the evidence on whether the reference specification is nsupported or not by the toxicological point of view.<br><br>Glyphosate reference specification:<br><br>The impurity glyphosine showed a potential for clastogenicity in an *in vitro* chromosomal aberration assay that was not appropriately followed up *in vivo*; however, this impurity was present in some of the batches used in toxicity studies at levels representative of the proposed reference specification. Both relevance assessment and its maximum content were open for this impurity (not concluded, i.e. data gap), whereas there was evidence that it was present in some of the batches used in toxicity studies at levels representative of the proposed reference specification. Therefore, this leads to an issue that could not be finalised and a critical area of concern is not deemed warranted.<br><br>Cypermethrin reference specification:<br><br>The impurity hexane in cypermetrhin reference specification is a relevant impurity (no data gap identified to address its relevance); however, its maximum content in the proposed specification was not supported (lack of knowledge about the detailed composition of the batches used in toxicity studies (e.g. at least for the critical studies)). The relevance of this impurity was concluded (i.e. relevant) but not its maximum content. This leads to |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | | a critical area of concern given its known relevance (EFSA, 2018[20]).<br><br>Isoflucypram:<br><br>The impurity N,N-dimethylcyclohexanamine (BCS-AA10447) in isoflucypram reference specification is a relevant impurity (no data gap identified to address its relevance); however, its maximum content in the proposed specification was not supported. The relevance of this impurity was concluded (i.e. relevant) but not its maximum content. This leads to a critical area of concern given its known relevance (EFSA, 2022[21]).<br><br>Benfluralin:<br><br>The impurity ethyl-butyl-nitrosamine in benfluralin reference specification is an impurity of known toxicological concern (no data gap identified to address its relevance); however, its maximum content in the proposed specification was not supported. The relevance of this impurity was concluded (i.e. relevant) but not its maximum |

[20] EFSA (European Food Safety Authority), Arena M, Auteri D, Barmaz S, Brancato A,Brocca D, Bura L, Carrasco Cabrera L, Chiusolo A, Civitella C, Court Marques D, Crivellente F,Ctverackova L, De Lentdecker C, Egsmose M, Erdos Z, Fait G, Ferreira L, Greco L, Ippolito A, Istace F,Jarrah S, Kardassi D, Leuschner R, Lostia A, Lythgo C, Magrans JO, Medina P, Mineo D, Miron I, Molnar T,Padovani L, Parra Morte JM, Pedersen R, Reich H, Sacchi A, Santos M, Serafimova R, Sharp R, Stanek A,Streissl F, Sturma J, Szentes C, Tarazona J, Terron A, Theobald A, Vagenende B, Van Dijk J and Villamar-Bouza L, 2018. Conclusion on the peer review of the pesticide risk assessment of the active substance cypermethrin. EFSA Journal 2018;16(8):5402, 28 pp. https://doi.org/10.2903/j.efsa.2018.5402

[21] EFSA (European Food Safety Authority), Alvarez F, Arena M, Auteri D, BinagliaM, Castoldi AF, Chiusolo A, Colagiorgi A, Colas M, Crivellente F, De Lentdecker C, Egsmose M, Fait G,Ferilli F, Gouliarmou V, Herrero Nogareda L, Ippolito A, Istace F, Jarrah S, Kardassi D, Kienzler A,Lanzoni A, Lava R, Leuschner R, Linguadoca A, Lythgo C, Magrans O, Mangas I, Miron I, Molnar T,Padovani L, Parra Morte JM, Serafimova R, Sharp R, Szentes C, Terron A, Theobald A, Tiramani M and Villamar-Bouza L, 2022. Conclusion on the peer review of the pesticide risk assessment of the active substance isoflucypram. EFSA Journal 2022;20(6):7328, 34 pp. https://doi.org/10.2903/j.efsa.2022.7328

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | | content. This leads to a critical area of concern given its known relevance. (EFSA, 2022)[22]. |
| 13. | Paragraphs 59-61<br><br>Predominant weight to industry studies | # 61. The assessment of the genotoxic potential of glyphosate clearly does not meet these requirements [*refer to paragraphs 59-60*]. The conclusions on the absence of genotoxicity are implausible in view of the overwhelming positive results observed in the independent scientific literature, including the most recent ones. This literature has been systematically excluded or subweighted for reasons that are neither legally nor scientifically founded. According to **EFSA's own recommendations,** positive results established *in vitro* should have led to more investigations, and in particular to an *in vivo* comet test. | All pertinent studies on genotoxicity were part of the hazard assessment undertaken in the context of the formal assessment of the proposal for harmonised classification and labelling in accordance with Regulation (EC) No 1272/2008 carried out by ECHA in parallel to the EFSA peer review, leading to the conclusions as delivered in the RAC Opinion on 30 May 2022 (ECHA, 2022).<br><br>In the EFSA peer review, all available studies, both from the applicants and from publications, have been duly considered and assessed for their relevance and reliability following a rigorous approach as detailed in the RAR. Where needed, additional information has been requested during the regulatory stop of the clock to complete the data package and address the comments received during the public and targeted MS consultation.<br><br>With regard to the genotoxicity assessment, contested in this and the preceding paragraphs, EFSA did identify several data requirements for the applicants, please refer to the Reporting table (section 2) points 2(149), 2(151), 2(152), 2(153), 2(154), 2(155), 2(158), 2(161), 2(165), 2(169), 2(201), 2(202), 2(203), 2(204) and Reporting table identified following public comments (section 2) points 2(43), 2(44), 2(51). |

[22] EFSA (European Food Safety Authority), Alvarez F, Arena M, Auteri D, Binaglia M,Castoldi AF, Chiusolo A, Colagiorgi A, Colas M, Crivellente F, De Lentdecker C, Egsmose M, Fait G, FerilliF, Gouliarmou V, Herrero Nogareda L, Ippolito A, Istace F, Jarrah S, Kardassi D, Kienzler A, Lanzoni A,Lava R, Leuschner R, Linguadoca A, Lythgo C, Magrans O, Mangas I, Miron I, Molnar T, Padovani L,Parra Morte JM, Rizzuto S, Serafimova R, Sharp R, Szentes C, Terron A, Theobald A, Tiramani M and Villamar-Bouza L, 2022. Conclusion on the peer review of the pesticide risk assessment of the active substance benfluralin. EFSA Journal 2022;20(9):7556, 28 pp. https://doi.org/10.2903/j.efsa.2022.7556

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | | In addition, a dedicated expert consultation on all relevant endpoints took place, including specific discussion on the impact of the identified deviations of the genotoxicity studies on glyphosate (considerations of major vs minor deviations). The experts' consultation resulted in the collegial agreement by the experts on the relevance and reliability of the studies, taking into account deviations from the applicable test guidelines (TGs).<br><br>The weight of evidence approach for genotoxicity on glyphosate during the peer review included more than 70 studies (regulatory and public literature studies) assessed as acceptable, supplementary or supportive (see Renewal Assessment Report at Table 2.0.5.2-3 and Table 2.0.5.2-4 (RAR, Volume 1, 2023)).<br><br>Overall, the genotoxicity data package on glyphosate was considered extensive and sufficiently robust that permit proper conclusions to be drawn on genotoxicity without the need for requesting further vertebrate studies.<br><br>The conclusion was in line with the RAC opinion. |
| 14. | Paragraphs 62-65<br><br>Manifest error in the genotoxicity assessment – lack of excellence and independence | # 62. In fact, the conclusions of the evaluation are mainly based on the tests carried out by the applicants for re-approval. However, these trials do not reflect the "*most recent results of international research*", let alone the principles of "*transparency, excellence and independence*". They (i) are almost all based on protocols that are obsolete and dating back several decades and/or are affected by other methodological limitations; (ii) they cover a single tissue; and (iii) they are characterised by a very low sensitivity. As for the UDS tests, they were themselves declared unreliable and irrelevant in the RAR in line with the OECD and EFSA positions, so that they could not be used to | All available studies, both from the applicants and from publications, have been duly considered and assessed for their relevance and reliability following a rigorous approach as detailed in the RAR. Where needed, additional information has been requested during the regulatory stop of the clock to complete the data package and address the comments received during the public and targeted MS consultation.<br><br>With regard to the genotoxicity assessment, contested in this and the preceding paragraphs, EFSA did identify several data requirements for the applicants, please refer to the Reporting table (section 2) points 2(149), 2(151), 2(152), 2(153), 2(154), 2(155), 2(158), 2(161), 2(165), |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | minimise the significance of positive results from *in vitro* tests reported in independent literature.<br><br># 64. The contrast is clear with the way in which the assessment of another active substance suspected of being genotoxic, namely chlorpyrifos-methyl, was conducted. In that dossier, the RAR states that ' *Although the two [articles] were not carried out in accordance with [good laboratory practice] and used new techniques not included in the standard guidelines, the concerns resulting from their results could not be rejected by the studies provided [by the applicants], since [the latter] did not allow for the analysis of a wider range of DNA alterations*"[92] This risk assessment has been performed in the right way: if the concerns from independent scientific literature cannot be addressed by the studies provided by the applicants, it must be concluded that the demonstration of the absence of adverse health effects is not provided. | 2(169), 2(201), 2(202), 2(203), 2(204) and Reporting table identified following public comments (section 2) points 2(43), 2(44), 2(51).<br><br>In addition, a dedicated expert consultation on all relevant endpoints took place, including specific discussion on the impact of the identified deviations of the genotoxicity studies on glyphosate (considerations of major vs minor deviations). The experts' consultation resulted in the collegial agreement by the experts on the relevance and reliability of the studies, taking into account deviations from the applicable test guidelines (TGs).<br><br>Overall, the genotoxicity data package on glyphosate was considered extensive and sufficiently robust that permit proper conclusions to be drawn on genotoxicity without the need for requesting further vertebrate studies.<br><br>As for glyphosate, also the assessment of chlorpyrifos considered studies from the applicants and from publications. However, the genotoxicity data package on chlorpyrifos was not as sufficiently robust as for glyphosate: "*the genotoxicity potential remained unclarified (positive findings from an in vitro chromosome aberration study and two in vitro unscheduled DNA synthesis assays; in vivo positive findings from open literature on chromosome aberration and on DNA damage caused through oxidative stress or by topoisomerase II inhibition which was considered a MIE for infant leukaemia* (EFSA statement on chlorpyrifos, 2019[23]). As regards chlorpyrifos-methyl, "*the available regulatory genotoxicity data set submitted for chlorpyrifos-methyl did not show any concern. The experts highlighted that very limited literature* |

[23] EFSA (European Food Safety Authority), 2019. Statement on the available outcomes of the human health assessment in the context of the pesticides peer review of the active substance chlorpyrifos. EFSA Journal 2019;17(8):5809, 23 pp. https://doi.org/10.2903/j.efsa.2019.5809

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | | *data were retrieved specifically for chlorpyrifos-methyl. Considering also the read-across discussion, most experts decided to precautionary apply to chlorpyrifos-methyl the same conclusions as for chlorpyrifos. Therefore, the experts concluded that the genotoxicity potential of chlorpyrifos-methyl remains as unclear as that of chlorpyrifos"* (EFSA statement on chlorpyrifos-methyl, 2019[24]). |
| 15. | Paragraphs 66-100<br><br>Illegalities and manifest errors in the assessment of the carcinogenicity of glyphosate | *Paragraphs linked to classification criteria and elements falling in the remit of ECHA*. | EFSA agrees on the conclusions reached in the ECHA RAC and by the assessment of the carcinogenicity studies and epidemiological data performed by the RMS in the RAR. |
| 16. | Paragraphs 101-115<br><br>Illegalities and manifest errors in the assessment of the effect of glyphosate on microbiome | # 103. In its Conclusion, however, EFSA states: Several studies from the published literature investigated the potential effects of glyphosate on the human and animal gut microbiome, and possible consequent effects on health. Based on the current state of knowledge, considering that standardised regulatory guidance and/or established harmonised criteria are currently not available for the assessment of microbiome, no definitive conclusions can be drawn from these studies. However, the available mammalian toxicity dataset supports a sufficiently protective assessment for any health impact possibly mediated by the microbiome on humans, livestock and pet animals. Consistently, the previous conclusions on the lack of impact of glyphosate on animal gut microbiome and | PAN claims that the EFSA assessment of glyphosate (# 103) would have not adequately taken into account the possible effects on the gut microbiome of humans and animals (and possible health consequences) due to the lack of guidelines/standardised methods (# 104), without an adequate search and evaluation of independent, relevant scientific and technical information (# 112, 113 and 114). This would equally apply to the active substance, its metabolites and impurities (# 106, 110 and 111). This approach would have led to contradictory conclusions (# 105), to breach of the legal requirements by the PPP regulation (# 106) and by the EFSA founding regulation regarding excellence and independency (# 107 and 115) and would imply a more cautious approach as regards conclusions on safety (# 109). Additionally, PAN considers |

[24] EFSA (European Food Safety Authority), 2019. Updated statement on the available outcomes of the human health assessment in the context of the pesticides peer review of the active substance chlorpyrifos-methyl. doi: 10.2903/j.efsa.2019.5908

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | health (EFSA, 2018a) valid. Further developments are required to understand the importance of the microbiome in risk assessment and identify dedicated strategies and methodologies accordingly (..)[119]<br><br># 104. In other words, EFSA considers that, in the absence of harmonised criteria or standardised guidelines, indications of the effect of glyphosate on the human microbiome can simply not influence the assessment of the risk of that substance to human and animal health. In the alternative, it considers that the other toxicity data have in any event led to a sufficiently protective assessment for any type of health impact potentially caused through the microbiota.<br><br># 105. This alternative reasoning is in no way substantiated. Moreover, it is fundamentally contradictory to argue both that it is impossible to carry out a risk assessment of the impact on microbiota and that such risks – the severity and extent of which are by definition unknown – are in any event already covered by the other data. The conclusions on neurotoxicity, for example, are mainly based on tests, the design of which makes them incapable of revealing a possible mode of action related to microbiota. Furthermore, microbiome alterations have effects on human health that go far beyond the toxicity areas examined in the RAR and the EFSA conclusions (increase in allergies and intolerances, alteration of the digestive system, etc.)[120,121].<br><br># 106. As regards the main statement of reasons, it fails to comply with the requirements of the PPP Regulation. In this regard, it should be noted that | that some studies should be followed up by additional investigations (# 113 and 114).<br><br>EFSA does not agree with the above PAN considerations.<br><br>It is a matter of fact that currently there are no specific regulatory requirements or guidance documents in place for plant protection products to specifically investigate possible effects on microbiomes or effects by microbiomes on human and animal health; this is common to other (food) regulatory areas, and substantially derives from a scientific backbone currently still insufficient to the purpose. Certainly, standardised protocols would facilitate the consistent and reliable assessment of the microbiome in the pesticide area, nevertheless EFSA agrees that the lack of standardized protocol *is not a sufficient argument* to dismiss possible effects on the gut microbiome, particularly considering possible antimicrobial properties of glyphosate.<br><br>In integration to the assessment and to fulfill the legal requirements a literature search has been carried out for glyphosate and its metabolites, with search strings including keywords relevant for investigating microbiome. During the peer review process, additional studies were identified. The set of publicly available studies on the gut microbiome, its perturbations and consequence for the health of humans and animals (livestock and pets) included primary research studies, reviews, commentaries, editorials and EFSA outputs, was considered potentially relevant and duly assessed by a dedicated working group (57 studies). All the studies indicated by PAN were assessed in the peer review process, but Chen et al 2021, Puigbo et al 2022 and Walsh et al 2023. The retrieved information provided an up-to-date overview of activities conducted to explore the possible effects of glyphosate on gut microbiomes in |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | Article 4 of the PPP Regulation only allows the approval of active substances which, '*in the light of current scientific and technical* knowledge', can be *expected to have 'no harmful effects on human health, including vulnerable groups, or on animal* health'. It follows from that provision that:<br>– Any harmful effects on human or animal health must lead to a refusal of approval;<br>– These harmful effects must be assessed in the light of current scientific knowledge.<br><br># 107. In other words, the absence of *ad hoc* guidelines or guidance documents cannot be used as an excuse for competent authorities to avoid the assessment of a health effect that is extensively documented in independent literature. As a reminder, according to EFSA's founding regulation, '*the risk assessment shall be based on the scientific evidence available*'[122] and it is for EFSA *to 'provide the Community institutions and the Member States with the best possible scientific advice*'[123], *which means 'to seek, collect, collate, analyse and summarise scientific and technical data in the fields falling within its mission*'[124]. Where appropriate, " *on the basis of the best available independent scientific sources, the Authority shall commission the scientific studies necessary for the fulfilment of its mission*"[125].<br><br># 108. It cannot be inferred from any provision of that regulation that an EFSA scientific opinion must necessarily be based on prior guidelines or standardised methods. On the contrary, it is up to EFSA, if necessary, | humans and domestic animals and possible consequent effects on health at the time of the assessment.<br><br>The retrieved information was judged of unclear relevance and overall not adequate to derive definitive conclusions on glyphosate effects on the gut microbiota for a series of reasons, including: studies were conducted according to variable, not standardised approaches using a variety of tools and methodologies, this making the comparability and repeatability of results difficult; such studies often aimed at investigating specific microbiota populations and/or specific hypothesis, not the whole gut microbiome and/or organisms' response; the causal links between the microbiome (and its dysfunctions) and humans/animals pathological conditions need further consolidation, as indicated by most authors; finally, various weaknesses were identified, hampering reliability. Interestingly, these considerations are aligned with the recent publication by Moreno et al, 2024 (see below).<br><br>The full information on the identified studies and methodology for their appraisal is reported in the supporting published documentation of the Pesticides Peer Review Experts' Meeting TC 80 (see Annex 9 to the Peer Review Meeting report[25]).<br><br>Regarding the new publications specifically mentioned by PAN, Puigbo et al., 2022 reports about in silico studies on potential targets for glyphosate on the human microbiome. The authors admit that this necessitates support by empirical studies and epidemiological investigations to clarify the effect of glyphosate on the healthy human microbiome. Chen et al 2021 and Walsh et al 2023 are |

[25] available in the Peer Review Report in the Open EFSA under 'Supporting documents' under EFSA Question number EFSA-Q-2020-00140: (https://open.efsa.europa.eu/study-inventory/EFSA-Q-2020-00140); refer to Part 3 – TC 80_Peer Review Report_Glyphosate_Annexes. Refer to Annex 9

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | to propose criteria or technical guidelines *on the basis of prior scientific data.* In other words, as Advocate General Medina has pointed out, ` *the obligation to apply the most recent scientific and technical knowledge, which requires the competent authority to be proactive by seeking to better protect human and animal health and the environment*'[126], even if it means ignoring the regulatory limits. Recent decisions of national courts go in the same direction[127].<br><br># 109. Furthermore, insofar as the PPP Regulation makes the approval of an active substance subject to proof of the absence of any harmful effects on health, evidence from independent scientific literature showing the presence of an effect of glyphosate on the composition and activity of microbiota should, in itself and pending a standardised method to characterise and specify the health risk, lead to a refusal of approval of that active substance under the precautionary principle. The fact that, as EFSA claims, 'no *definitive conclusion can be drawn*'[128] in the absence of guidelines justified a cautious approach. It is completely contrary to the spirit of the PPP Regulation and Article 1 (4) thereof to impose on human health a risk linked to a failure on the part of EFSA and the Member States*, a* fortiori where that risk concerns cut-off criteria such as reproductive toxicity, which includes developmental neurotoxicity[129].<br><br># 111. The applicants for re-approval clearly did not comply with these requirements (*see # 110 in the review request*) as regards the information on the | reviews about respectively possible effect that glyphosate has on the human body with a specific focus on the gut microbiome and the potential consequences for human health, and on the regulatory mechanisms of the intestinal microbiome and its metabolites (mainly neurotransmitters and their precursors) on cognitive functions and the pathogeneses of neurodegenerative diseases such as Alzheimer and Parkinson disease. Overall, these studies do not add evidence that change the current conclusions on glyphosate.<br><br>In the absence of definitive information from the open literature, EFSA reiterates that the current assessment of glyphosate was based on a robust, up-to-standard data package and the derived current toxicological reference values are considered protective towards all the observed adverse effects, including those that could be secondary to gut microbiome perturbation, under the current state of knowledge.<br><br>As regards proactivity, beside the considerations on the literature search above, EFSA acknowledges that the field of microbiome research has evolved rapidly over the last years and could play an important role in various areas of EFSA's scientific assessments. In June 2020, EFSA published an editorial (Merten et al, 2020)[26], highlighting that gut microbiome research is expected to play a relevant role in regulatory science and that further research is needed to enhance the understanding of the toxicological significance of microbiome-mediated metabolism of chemicals. To start building this capacity, EFSA launched a thematic grant in March 2020 (GP/EFSA/ENCO/2020/02) on this topic to collaborate with EU Member States and to |

[26] Merten C, Schoonjans R, Di Gioia D, Pelaez C, Sanz Y, Maurici D, Robinson T, 2020. Editorial: Exploring the need to include microbiomes into EFSA's scientific assessments. EFSA Journal 2020;18(6):e18061, 7 pp. https://doi.org/10.2903/j.efsa.2020.e18061

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | effects of glyphosate on microbiota, which should have led the RMS to declare their dossier inadmissible.<br><br># 112. First, their state of scientific literature on the issue is deficient. It is far from covering the approximately 150 studies, which, according to PubMed[131], deal with the relationship between glyphosate and microbiome.<br><br>#113. Second, in view of the results and analyses provided in the independent literature, the applicants for re-approval should have conducted tests capable of confirming or refuting them. This work was clearly not undertaken – or, at the very least, the results of that work were not forwarded to the competent authorities. Only one study, which did not concern microbiome analysis, produces interpellant results: it was concluded that an exacerbation of anxiety and depression-like behaviour and significant disturbances in relative abundance and phylogenic diversity of gut microbiota in mice were induced by GBH exposure[132.]<br><br>#114. However, the RMS considered that such a study was "not directly relevant" since the intestinal microbiota study "*is currently not part of the European assessment framework for pesticides*"[133]. For the same reason, it is apparent from Volume 1 of the RAR that no independent literature study on the subject, including | identify indications for future EU research agendas with a focus on specific needs from a risk assessment perspective; reports have been published in February 2024 (Moreno et al, 2024 [27] and Debode et al, 2024 [28] ). As regards mammalian gut microbiome (Moreno et al, 2024) the outcome of the work converges with EFSA's conclusions on glyphosate, indicating a lack of consistency and standardisation in methodologies necessary for robust comparison both in humans and animals, and a major lack of understanding of the underlying molecular mechanisms mediated by the gut microbiome and their link to host phenotypes, as well as dose-dependent effects. This applies to glyphosate and its metabolite AMPA, also investigated in the literature search run by Moreno et al, 2024. This work proposes a roadmap for future activities of relevance for incorporating the assessment of microbiomes in risk assessment is proposed, as well as multidisciplinary research strategy to provide key information to fill knowledge and methodology gaps and eventually developing policy actions aiming at the elaboration of decision frameworks for the future incorporation of gut microbiome data into specific guidelines and, ultimately, into regulatory programmes. |

[27] Moreno, Francisco Javier; Florencio Pazos, Manuel Garrido-Romero, Cyrielle Payen, Gonzalo Borrego-Yaniz, Mónica Chagoyen, Nieves Corzo, Martine Denis, Christelle Fablet, María Fernández, Adela Granja, Maryse Guinebretière, Muriel Guyard, Rodrigo Jiménez-Saiz, Alassane Keita, Annaëlle Kerouanton, Ana Márquez, Javier Martín, Antonia Montilla, Ana Muñoz-Labrador, Jorge Novoa, Frédéric Paboeuf, Marta G. Rivera-Ferre, Patricia Ruas-Madiedo, Lorena Ruiz, Amandine Thépault, Mar Villamiel, Carlos Benito and Marianne Chemaly, 2024. Roadmap for the integration of gastro-intestinal (GI) tract microbiomes (human and domestic animal) in risk assessments under EFSA's remit. EFSA supporting publication 2024:EN-8597. 238 pp. doi:10.2903/sp.efsa.2024.EN-8597

[28] Debode F, Caulier S, Demeter S, Dubois B, Gelhay V, HulinJ, Muhovski Y, Ninane V, Rousseau G, and Bragard C, 2024. Roadmap for the integration of environmental microbiomes in risk assessments under EFSA's remit. EFSA supporting publication 2024:EN-8602.93pp. doi:10.2903/sp.efsa.2024.EN-8602

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | those submitted in the public consultation, could be taken into account in the weight-of-evidence assessment[134].<br><br># 115. This way of excluding independent literature on the grounds that it does not comply with hypothetical regulatory standards that do not exist again shows serious breaches of the standards of excellence and independence that are supposed to govern risk assessment. | |
| 17. | Paragraphs 116-119<br><br>Illegalities and manifest errors in the assessment of the effect of glyphosate on microbiome – non target organisms | # 116. The same applies to the assessment of environmental toxicity. It follows from the EFSA Conclusion that:<br>For the current assessment, studies were identified (both via literature search and submitted during the consultation phase) on the potential effects of glyphosate and formulations on the microbiome of non-target organisms. The information was assessed for relevance and reliability using criteria agreed during the Pesticides Peer Review Experts' TC 82. The impact of glyphosate on the microbiome was discussed at the Pesticides Peer Review Experts' TC 82 and also at the Pesticides Peer Review Experts' TC 80 on mammalian toxicology. Only for bees, the studies identified were evaluated as relevant and reliable and responses due to glyphosate exposure on bees' gut microbiota identified, such as changes in the abundance of core microbial species. In particular, a decrease in abundance and growth of bee gut bacterium Snodgrasella alvi was observed. Generally, it | PAN EU claims that the EFSA assessment of glyphosate on microbiota was limited to the potential effects on gut microbiome of bees (#117 and #118) and some publications for other non-target organisms were quoted in order to demonstrate that this information would have been available for a more comprehensive assessment. This allegation is not valid, as a significant amount of literature was taken into consideration to assess such potential effects on different non-target organisms. Only the studies assessed as relevant and reliable were further used (the criteria for evaluating relevance and reliability were agreed at the Pesticides Peer Review Experts' TC 82).<br><br>The quoted studies by Owagboriaye, 2021 and Ruuskanen, 2020 were included in the assessment. Bellot, 2023 and DeBeer *et al*. 2023 were published after the time frame for which the assessment of the literature was performed.<br><br>As regards Anderson 2017 and Rayman & Moran 2018, they are not primary research studies and they do not focus on pesticides (glyphosate is even not mentioned in these papers). It should be noted that the EFSA Conclusion does not claim that link between dysbiosis of individual bees and effects on population does not exist; EFSA's Conclusion only flags that the relevance of such effects is not known. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | was acknowledged that the relevance of these effects at the population level is unknown[135].<br><br># 117. In other words, the abundant literature showing an effect on the microbiota of non-target species such as earthworms (Owagboriaye, 2021[136]), birds (Ruuskanen, 2020[137]) or fish (Bellot, 2023[138]) **was simply not taken into account**. Only studies on the effect of glyphosate on the intestinal microbiota of bees were considered reliable and relevant. Even on this point, however, EFSA merely pointed out that there was a lack of data on the impact of this effect on bee populations without drawing appropriate conclusions in the light of the requirements of the PPP Regulation. However, other scientific literature studies indicate the importance of intestinal microbiota on bee health. In particular, Anderson (2017)[139] summarizes all scientifically demonstrated roles of the gut microbiota of the bee: development and growth, immunity, nutrition and effect on insulin levels are among the vital functions directly related to microbiota. Other publications (Rayman & Moran 2018[140], DeBeer *et al*. 2023[141]) link honeybee microbiota to susceptibility to pathogens or xenobiotic agents. It should be noted that, under point 3.8.3 of Annex II to the PPP Regulation, an active substance can only be approved if it is demonstrated that under the proposed conditions of use it " *will lead to negligible exposure of bees*" or " *will not have unacceptable acute or chronic effects on colony survival and development, taking into account the effects on honeybee larvae and bee behaviour*". In particular, it is stipulated that no substance likely to lead to a reduction of more than 10 % in honeybee populations does not meet those criteria[142]. In view of | PAN EU claims that an attempt should have been made to quantify the effects on the population (in order to assess whether a reduction of more than 10 % in colony strength may happen). Indeed, the applicant was not requested to make such an attempt, however it was clear for EFSA that such method does not exist and, in general, this scientific topic needs further research and development. Although the quoted publications that investigated the importance of such effects (Anderson 2017 and Rayman & Moran 2018, DeBeer *et al*. 2023) were not assessed, they seem to only reiterate that effects on individual level may escalate to higher organisation level.<br><br>EFSA disagrees with the claim (#118) that there is ample evidence from independent scientific literature demonstrating that the health impact on honey bees has such importance that it should have led to the conclusion that unacceptable effects were demonstrated (which would have needed to be demonstrated for the colony). Therefore, EFSA disagrees with the claim (#119) that the burden of proof was reversed. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | these criteria and considering the established effect of glyphosate on honeybee microbiota and its importance on bee health, EFSA (or RMS) should have required from the applicants additional data to quantify and characterise the risk to bees. Failing that, it should have considered that the applicants had not demonstrated that those criteria were not met, and that glyphosate could therefore not be re-approved.<br><br># 118. In conclusion, the risk assessment of a possible alteration of the microbiome was simply not carried out. However, there is ample evidence from independent scientific literature, relating both to *(i)* the effect of glyphosate on the composition of the microbiome and *(ii)* to the health impact from the alteration of the microbiome. In the presence of those studies, and in the absence of a regulatory study capable of putting them into question, EFSA had no choice but to conclude that the absence of adverse effects on human and animal health and the absence of unacceptable effects on the environment had not been demonstrated. As a reminder, "in order *for the application to be rejected, i.e., for a measure to be adopted which both restricts the rights of the producer applying for renewal of the approval of an active substance and protects human health, it is sufficient that mere uncertainty as to the presence of a risk concerning that substance can be identified*"[143].<br><br># 119. Contrary to this conclusion, EFSA considered that, in the absence of criteria and technical guidelines on this issue, the effects caused by glyphosate on the microbiome could not be assessed, and therefore could not lead to a finding of non-compliance with the criteria | |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | of Article 4 of the PPP Regulation. By this reasoning, making the risk assessment conditional to the drafting of guidelines and their subsequent adoption by political bodies (the Commission and the Member States), EFSA is reversing the burden of proof and undermines the whole logic of the risk assessment and management system established by the PPP Regulation and, more broadly, by Regulation 178/2002. | |
| 18. | Paragraphs 122 to 131<br><br>Unlawfulness and manifest errors in neurotoxicity assessment<br><br>(a) Neurotoxic effects from independent scientific literature | # 122. The neurotoxicological effects of glyphosate are established by numerous studies from independent literature. However, this was not properly identified and examined (a).<br>Moreover, the only developmental neurotoxicity study carried out on a glyphosate salt, which proved to be positive, was not submitted by the applicants for re-approval, and was excluded without serious scientific reasoning (b). The studies submitted by the applicants for re-approval are insufficient to establish the absence of developmental neurotoxicity (c). Finally, the EFSA Conclusion does not properly reflect the concerns that emerged from the review of the dossier (d).<br><br><u>(a) Neurotoxic effects from independent scientific literature</u><br><br># 123. A systematic review of scientific literature from the last 10 years indicates that exposure to glyphosate or its commercial formulations causes several neurotoxic effects in humans and other species (Costas-Ferreira *et al.,* 2022)[146], a conclusion also drawn by INSERM (2021)[147]. Of the 921 articles identified on glyphosate and neurotoxicity, 51 were selected following strict criteria to ensure their relevance for the | #122 and #123. The set of studies considered in the renewal assessment of glyphosate included a package of neurotoxicity studies performed in rodents (one acute and two sub-chronic neurotoxicity studies in rats) and a delayed polyneuropathy study (one delayed neurotoxicity study in domestic hens), in agreement with the data requirement as set out in Commission Regulation (EU) No 283/2013. As indicated in the EFSA Conclusion (2023), no indication of neurotoxicity potential of glyphosate was present from the above-mentioned studies. In addition, a systematic literature review has been submitted by the applicants via the required updated literature review of the last 10 years preceding the dossier submission (from January 2010 until end of December 2019 and extended up to end of June 2020) in line with the legal requirements and the peer review process. All public literature submitted to EFSA throughout the regulatory process for the renewal of glyphosate (i.e. included in the RAR or requested during the public commenting phase) was considered as potentially relevant and included in the assessment. Additional literature identified after the public consultation and considered appropriate to support the assessment was also included. Public literature available to EFSA included primary research studies (in vivo, in vitro and mechanistic), epidemiological studies, reviews, etc. The documents used |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | assessment, of which 23 concerned tests on the active substance glyphosate (and not on its formulations). The review indicates that glyphosate has the capacity to pass the blood-brain barrier (Martinez *et al.,* 2019[148]), which could partly explain the different types of short-term or long-term disturbance that have been observed in the human nervous system. Several animal studies (16 *in vivo* and 6 *in vitro*) show that glyphosate or glyphosate-based herbicides have a range of toxic effects on the central and peripheral nervous system. The main effects observed include changes in the development of the nervous system and neurotransmission systems, as well as oxidative stress and neuroinflammation, leading to neurons death and behavioural changes. As stated in the journal, "*Most of these studies show the neurotoxic effects of glyphosate administered at early ages during the intrauterine period and lactation, although chronic or acute exposure in adulthood also causes important alterations in the function and structure of the nervous system*". The authors also examined studies on fish. In line with the observations made in rodents, the 21 studies analysed (including 9 on the active substance alone) showed that glyphosate mainly affects the development of the nervous system, neurotransmission, behaviour and energy metabolism, while causing oxidative stress and inflammation.<br><br># 124. Recent studies provide further evidence of neurotoxicity of glyphosate and glyphosate based herbicides. In terms of developmental neurotoxicity, the early exposure of rats to glyphosate (postnatal day | during the commenting phase, the RAR and its revised version were used as reference documents. To assess the available literature on neurotoxicity, EFSA with the support of a Working Group followed a structured approach as described in the Annexes 7 (neurotoxicity studies) and 4 (epidemiological studies, including neurotoxicity) of the Pesticides Peer Review Experts' Meeting Report TC 80[29]. The scope of the activity was to assess the available information and provide a weight-of-evidence evaluation of the possible effects of glyphosate on human health. Individual studies commented during the public consultation phase were grouped in different sub-sections (e.g., autism, Parkinson's disease, neurotransmitters, developmental neurotoxicity and other neurotoxicity studies). Overall, the evidence made available to the EFSA Working Group on neurotoxicity (epidemiological studies included) was limited and quite heterogeneous regarding exposure (most studies used GBH rather than glyphosate active substance, and at a very wide dose range) as well as the endpoints assessed. In general, the reliability of the studies was considered low to infer causal associations, which limited their utility for risk assessment as no robust conclusion could be drawn. As previously indicated, the additional papers identified after public consultation and considered appropriate to support the assessment were further assessed and the outcome included in Annex 4 and 7 of the Pesticides Peer Review Experts' Meeting Report TC 80.<br><br>#124. Regarding developmental neurotoxicity, a DNT study was not performed with glyphosate acid and considered not needed in the absence of neurotoxic effects in the |

[29] available in the Peer Review Report in the Open EFSA under 'Supporting documents' under EFSA Question number EFSA-Q-2020-00140: https://open.efsa.europa.eu/study-inventory/EFSA-Q-2020-00140); refer to Part 3_Peer Review Report_Glyphosate_Annexes. TC 80. Refer to Annexes 4 and 7.

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | 7 to 27, via subcutaneous injection) led to cognitive impairments, as spatial recognition and memory were altered, due to observed impacts of glyphosate on synapses and neural connectivity (Luna *et al.*, 2021[149]). In a previous study of similar design, gestational exposure to glyphosate has led to changes in reflex development, motor activity and cognitive function in new-born rats, in a dose-dependent manner, and altered a key neural signalling route, involved in the development and maturation of neurons (Coullery *et al.*, 2020[150]). In another study, maternal exposure to a product containing glyphosate in the form of isopropylamine salt (IPA) – potentially the representative formulation – led to numerous behavioural and cognitive abnormalities in offspring, closely associated with significant histological, neurochemical and molecular alterations (Ait-Bali *et al.*, 2020[151]). In another study, the exposure of rats to glyphosate IPA salt or to a glyphosate-containing formulation during early development from the 6th day of gestation to postnatal day 21 had similar effects on neurogenesis disturbance, and these effects continued until adulthood, accompanied by compensatory responses and the induction of oxidative stress in the brain hippocamp region (Ojiro *et al.*, 2023[152]).<br><br># 125. An epidemiological study carried out in California in the United States (von Ehrenstein *et al.*, 2019[153]) examined 2961 persons diagnosed with autism spectrum disorders to determine prenatal and infant exposure to pesticides. Glyphosate was one of the pesticides to which exposure during the prenatal period was associated with a risk of autism spectrum disorder, and it was the one which showed the highest | regulatory data set available for glyphosate. From the body of evidence assessed by the EFSA Working Group from a total of 7 literature studies (see Annex 7), although some effects were reported for GBH (commercial formulations) (Ait Bali et al., 2020) and various types of glyphosate salts (monoisopropylamine in the case of the study by Luna et al., 2021 and Coullery et al., 2020; and trimesium for the 2001 study), EFSA concluded that there was no clear pattern of effects suggesting DNT liabilities for glyphosate acid. Therefore, EFSA concluded that studies not conducted with glyphosate acid were not considered adequate to conclude on DNT while all the available evidence from studies conducted with glyphosate acid does not suggest any concern for DNT. A summary of the DNT study conducted with GBHs or alternative salts is reported in the minutes of the Pesticides Peer Review Experts' TC 80 and its Annex 7.<br><br>#125. From the 7 epidemiological studies available (see Annex 4), including reviews, and investigating the possible relationship between exposure to glyphosate and autism, only one found significant associations with glyphosate exposure (von Ehrenstein et al., 2019). However, it was not possible to assume unambiguous levels of exposure to single pesticides considering concomitant co-exposures, and the EFSA Working Group considered that no conclusion could be drawn on the possible correlation between exposure to glyphosate and autism since this unique study had limitations regarding exposure assessment. For the assessment of Ongono et al., 2020 (systematic review of epidemiological data in children and in vivo studies in rodents), Pu et al., 2020a, please see Annex 4 and Annex 7. The paper by Pu et al., 2021) was not considered by the Working Group, since being out of the time frame used for |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | association for autism spectrum disorder with intellectual impairment, in fact just above the association observed for chlorpyrifos, already known to affect children's brain development, and for which the Commission very logically adopted a non-re-approval decision[154]. Such epidemiological data were also used to support the non-re-approval of chlopyrifos-methyl[155], even though doubts remained as to the presence of chlopyrifos-methyl in detected organophosphates[156]. A systematic review of the association between early exposure to pesticides used in Europe and autism spectrum disorders showed that glyphosate had a ' *moderate level of proof*' in children, while in rats there was a ' *high level of evidence*' as to its association with the impairment of behavioural, learning and memory capabilities (Ongono *et al.*, 2020[157]). Behavioural abnormalities of ASD (for 'autism spectrum disorders') were observed in young mice after maternal exposure to high levels of a glyphosate-based herbicide (Pu *et al.*, 2020a[158]), as well as low doses of glyphosate alone (Pu *et al.,* 2021[159]), suggesting that it is glyphosate rather than the formulation that contributes to ASD behavioural abnormalities in juvenile offspring.<br><br># 126. An epidemiological study carried out in Washington State in the United States (Caballero *et al.*, 2018[160]) examined 4591 Parkinson's disease deaths between 2011 and 2015, of which 659 died prematurely and lived less than 1 000 metres in pesticide use areas. Glyphosate proved to be one of the two pesticides associated with premature mortality from Parkinson's disease, the other being paraquat, which was banned in Europe due to its high toxicity. Scientific literature reports specific cases of people exposed to glyphosate | the literature review and not identified after public consultation.<br><br>#126. Regarding the epidemiological studies on Parkinson's disease, 8 studies were made available to the EFSA Working Group and only Caballero et al., 2018 was considered acceptable with restrictions. This study found a significant association with glyphosate exposure, but the limitations inherent to GIS-based exposure assessment prevented from drawing robust conclusions. The remaining studies were case reports (Barbosa *et al.* 2001; Zheng *et al*. 2018; Wang et al., 2011), reviews or assessed exposure using an ecological approach (at the group level rather than at individual level). The study by Eriguchi et al., 2019 was not part of the studies assessed in the RAR, neither was it highlighted during the public consultation. EFSA notes that it is a case report of a 38-year-old man who developed parkinsonism 4 years after ingesting glyphosate. Regarding the study by Pu et al., 2020b, this was not considered since not part of the literature review and not identified after public consultation.<br><br>#127: Regarding amyotrophic lateral sclerosis (ALS) only two studies were made available with inconsistent results. While the AHS agriculture cohort found no association between glyphosate and ALS (Kamel et al., 2012), a case-control study (Andrew et al., 2021) reported a significant increased risk based on glyphosate exposure using GIS data (an indirect estimate of actual exposure that pose risk of misclassification). The study by Anderson et al., 2023 was not considered by the Working Group since being out of the time frame used for the literature review and not identified after public consultation.<br><br>#128: Regarding the potential effects of glyphosate on the human and animal gut microbiome, please see the outcome |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | and who have developed Parkinson's disease (Barbosa *et al.* 2001[161]; Zheng *et al.* 2018[162]; Eriguchi *et al.,* 2019[163]). Finally, in an animal experiment, where mice have been exposed to low levels of glyphosate for 2 weeks in combination with neurotoxin (MPTP, standard compound for Parkinson models in rodents), the combination has proven to be an additional neurotoxicant to the brain region known to be affected by Parkinson's disease (Pu *et al.*, 2020b[164]).<br><br># 127. Exposure to glyphosate was also associated with the development of amyotrophic lateral sclerosis (ALS) among residents of agricultural areas (Andrew *et al.*, 2021[165]). A recent study examining all available evidence suggests that the increased occupational risk of ALS among farmers, gardeners and sportsmen is closely linked to exposure during physical activity to glyphosate-based herbicides (Anderson., 2023[166]). The authors suggest that intestinal dysbiosis/permeability is one of the factors causing the disease.<br><br># 128. In recent years, the microbiome-intestine-brain axis has been considered an important factor in several neurological disorders. A review of the interaction between pesticide exposure and microbiome in the context of Parkinson's disease shows how changes in the human microbiome can cause such neurotoxicity (Kulcsarova *et al.*, 2023[167]). In 2019, a review of the impact of glyphosate on the intestinal microbiome and potential neurological effects revealed that glyphosate causes intestinal dysbiosis in beneficial bacteria | of the Pesticides Peer Review Experts' Meeting Report TC 80[30].<br><br>#129: As indicated under #123, all public literature submitted to EFSA throughout the regulatory process for the renewal of glyphosate (i.e. included in the RAR or requested during the public commenting phase) was considered as potentially relevant and included in the assessment. Additional literature identified after public consultation and considered appropriate to support the assessment was also included. These resulted in about 60 neurotoxicity studies considered (epidemiological studies included).<br><br>#130: As previously indicated, the available literature on neurotoxicity was assessed by the EFSA Working Group following a structured approach as described in the Annexes 7 (neurotoxicity studies) and 4 (epidemiological studies, including neurotoxicity) of the Pesticides Peer Review Experts' Meeting Report TC 80. It is noted that the study by Kamel et al., 2012 was assessed by the EFSA Working Group and not Kamel et al., 2007, as the latter was falling prior to the 10-year period before submission of the dossier. Studies by Luna et al., 2021 and Coullery et al., 2020 were assessed (see Annex 7 of the Peer Review Experts' Meeting Report TC 80) and discussed during the Experts' meeting TC 80. Regarding the potential effects of glyphosate on the human and animal gut microbiome, please see the outcome of the EFSA Working Group dealing with this topic and included in Annex 9 of the Peer Review Experts' Meeting Report TC 80. |

[30] available in the Peer Review Report in the Open EFSA under 'Supporting documents' under EFSA Question number EFSA-Q-2020-00140: https://open.efsa.europa.eu/study-inventory/EFSA-Q-2020-00140); also refer to Part 3_Peer Review Report_Glyphosate_Annexes. TC 80

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | populations. This has had an impact on the central nervous system and was associated with other neurobiological disorders due to the intestine-brain axis (Rueda-Ruzafa *et al.*, 2019[168]). These observations confirm previous concerns about the neurological impact of glyphosate, as it is an antibiotic agent and therefore exposure leads to progressive intestinal dysbiosis (Samsel and Sennef., 2015[169]). The conclusions of Inserm (2021) go in the same direction[170].<br><br># 129. This independent literature is roughly absent from the renewal dossier, which contains only 4 neurotoxicity studies – and even omits several studies listed in the previous renewal. It was therefore necessary to wait until the public consultation stage for at least 50 independent and published studies to be brought to the attention of the competent authorities, including 20 which had never been submitted before, within a timeframe, that made their assessment difficult[171] (**see Annex 6**).<br><br># 130. Of these 50 studies, only three were considered acceptable and fully incorporated into the risk assessment, one of which was before the 10-year period (Kamel *et al.*, 2007[172]) and the other was not peer reviewed (US EPA toxcast). All other studies were considered unacceptable or simply ` *supplemental*. As in the other sections (see *above*, point 130 on genotoxicity) many studies were excluded because they had been carried out on a glyphosate-based formulation and not on glyphosate itself, without any effort being made to ascertain the identity of the formulation, and assuming that the effects found are | # 131: The paper by Pu et al., 2021 was not considered by the Working Group since being outside of the time frame used for the literature review and not identified after public consultation. The studies by Martinez et al., 2018 and Martinez et al., 2019 were considered by the EFSA Working Group: Martinez et al., 2019 showed effects of glyphosate on the integrity of the blood brain barrier (BBB) at high doses in an in vitro study; Martinez et al., 2018 showed changes in dopaminergic neurotransmitters, particularly dopamine, but these effects were not observed at doses below 35 mg/kg bw per day for 6 days in this in vivo study in rats, and humans are highly unlikely to be exposed to higher dose (see Annex 7 of the Pesticide Peer Review Experts' TC 80). |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | due to the co-formulants. Similarly, many studies were underweighted on the grounds that they deviated from the OECD TG, while other – mostly negative – studies affected by the same 'defect' were considered relevant and reliable[173]. It should be added that, for the – illegal – reasons set out *above (see above*, paragraphs 101-119), microbiome studies have all been discarded, as well as studies on other species that could indicate common modes of action. Finally, it appears from the experts' meeting that some key studies were not even assessed by them at all, as they were not brought to their attention until the last minute[174].<br><br># 131. Ultimately, studies relating to neurotoxic effects of glyphosate played no role in the weight of evidence analysis, including studies considered relevant and acceptable such as those of Martinez *et al.* (2018[175] and 2019[176]) demonstrating that glyphosate crosses the blood-brain barrier and affects signalling pathways[177]. Even worse, some key studies have not even been identified and reviewed, including that of Pu *et al.* (2021)[178], reporting autism spectrum disorders in the offspring of mice exposed to the active substance glyphosate (and not to one of its formulations) at doses below the no-observed adverse effect dose (NOAEL). | |
| 19. | Paragraphs 132 to 145<br><br>Unlawfulness and manifest errors in neurotoxicity assessment | (b) Failure to take into account the only developmental neurotoxicity (DNT) study on glyphosate<br><br># 132. In a recent article, Mie and Ruden (2022)[179] reported that a DNT study was carried out in 2001 on the trimethylsulfonium salt of glyphosate, also known as sulfosate or glyphosate trimesium salt (Glyphosate-TMS). Although the contracted laboratory concluded | # 132: see also #124. The DNT study on glyphosate trimesium performed in 2001 was discussed during the Pesticides Peer Review Experts Meeting TC 80. Decreased motor activity and effects on learning and memory were observed in foetuses and not associated with maternal toxicity. ECHA considered this study in their RAC Opinion and concluded that, considering the low purity of the tested a.s. and missing information on its impurities, it was |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | (b) Failure to take into account the only developmental neurotoxicity (DNT) study on glyphosate | that the adverse effects observed in the DNT study were accidental (not related to treatment), EPA found an error in the interpretation of the results when assessing this study. It therefore concluded that this DNT study demonstrated behavioural effects on the offspring of rats after maternal exposure to Glyphosate-TMS during pregnancy and breastfeeding.<br><br># 133. Glyphosate, or N- (phosphonomethyl) glycine, is present in various chemical forms (salts or acid) in glyphosate-based formulations. In 2002, the European Commission included, inter alia, glyphosate and glyphosate-trimesium in its evaluation report for the active substance glyphosate[180]. The 2023 renewal procedure included glyphosate, its potassium, isopropylammine, ammonium and dimethylammonium salts, but not the trimesium salt.<br><br># 134. Although Glyphosate-TMS salt dissociates completely in water in glyphosate and trimesium and although effects have been observed on the offspring of exposed rats, this DNT study was not submitted by the applicants for re-approval. It should be noted, however, that the owner of the study, Syngenta, joined the consortium for the submission of the previous application in 2012, and subsequently remained a member of the consortium. Therefore, at least one of the applicants for re-approval was aware of this DNT study.<br><br># 135. It is true that Glyphosate-TMS is not included in the application for re-approval, unlike other glyphosate salts. The inclusion of the DNT study in the application was, however, an obligation under point 5.6 of the | difficult to assess if the effects reported were related to glyphosate trimesium or to the impurities. The data were evaluated also by US EPA and other deviations were mentioned: inadequacies in the assessment of learning and memory in the offspring and positive control data that could not be verified. Glyphosate trimesium was agreed to be a substance with a different toxicological profile than glyphosate acid. In the absence of a DNT study performed with glyphosate acid, and considering all the available evidence from the studies assessed by the EFSA Working Group and discussed also at the Pesticides Peer Review Experts' Meeting TC 80, EFSA identified a data gap to further investigate the cause of the observed effects from studies conducted with both GBHs and trimesium.<br><br># 133: During the 2023 renewal procedure, glyphosate (as isopropylammonium salt) was supported.<br><br># 134: As also clarified in # 140, the study was provided by the applicants following the specific request from EFSA, once EFSA has been made aware of the availability of this guideline DNT study on glyphosate-trimesium. The study was subsequently made available to both EFSA and ECHA as well as the RMS to allow consideration of all available evidence in the evaluations. During the Pesticides Peer Review Experts' Meeting TC 80 a specific action point was set for the RMS to include the assessment of the study performed with glyphosate-trimesium in a revised RAR.<br><br># 135: As reported in the minutes of the Pesticides Peer Review Experts' TC 80 and its Annex 7, glyphosate-trimesium is not considered a structural analogue of glyphosate acid, but a substance with evidence of |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | Annex to Implementing Regulation 283/2013, according to which " *analyses shall take into account all available and relevant data, including (...) knowledge on structural analogues of the active substance*". More generally, its inclusion was required under paragraphs 1.1, 1.2 and 1.3 of the introduction to Annex[181]. This is because;<br>– This DNT study is the only one available for glyphosate or one of its salts;<br>– " *the Court has stated, as regards the read-across method, that it is a method for the evaluation of substances widely recognised by the scientific community*' [182];<br>– That method is justified where ' *the two substances in question belong to the same group of chemicals* (...) *and, overall, have a similar chemical structure*'[183];<br>– This study should be considered relevant until proven otherwise, i.e. until the adverse effects reported are demonstrated to be related to trimesium salt itself or to the combination of trimesium salt and glyphosate and not to glyphosate. However, neither published scientific literature nor any studies submitted by applicants for re-approval provide information on such adverse reactions associated with trimesium salt;<br><br># 136. This omission has significant regulatory consequences. | neurotoxicity likely triggered by the trimesium ion (see US EPA assessment 2005)[31].<br><br># 136: Noted. EFSA does not have any specific comment on the point raised.<br><br># 137: During the Pesticides Peer Review Experts' Meeting TC 80 it was discussed if an additional uncertainty factor was needed in the derivation of reference values to cover the uncertainties related to the missing DNT study with glyphosate acid. However, no neurotoxic effects were observed in the available test guideline studies on glyphosate acid that would have triggered the need of an in vivo DNT study. Some evidence of DNT effects was present in studies conducted with different salts, different formulated products and non-appropriate routes of administration. Further assessment of the study by Ojiro et al., 2023 and the data from US EPA ToxCast/Tox21 Dashboard highlighted during the Pesticides Peer Review Experts' TC 80 confirmed that the reference values proposed are sufficiently conservative to cover the uncertainty related to the missing in vivo DNT study with glyphosate acid (see minutes of the Pesticides Peer Review Experts' TC 80 containing also a post meeting note discussing the assessment of the Ojiro et al., 2023 study and US EPA ToxCast/Tox21 Dashboard data)[32].<br><br># 138: Please see reply under #132<br><br># 139: Please note that in 2022, the ECHA RAC Committee (ECHA, 2022) concluded that no classification is warranted |

[31] U.S. EPA. *Data evaluation Record. Glyphosate Trimesium. Study type: developmental neurotoxicity study - rat; MRID 45539801.* 2005

[32] available in the Peer Review Report in the Open EFSA under 'Supporting documents' under EFSA Question number EFSA-Q-2020-00140: https://open.efsa.europa.eu/study-inventory/EFSA-Q-2020-00140); refer to Part 3_Peer Review Report_Glyphosate_expert meeting reports_public.pdf (TC 80, Expert consultation point 2.27)

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | # 137. On the one hand, under **point 3.6.1 of Annex II to the PPP Regulation, an increased margin of safety must be considered in order to set the acceptable daily intake (ADI) in cases of developmental neurotoxic or immunotoxic effects.** In other words, taking this study into account should have led to the establishment of an ADI lower than that laid down in the Renewal Regulation, as was the case for, for example[184].<br><br># 138. This assumption is far from theoretical. As Mie and Ruden (2022)[185] explain, in the DNT study the doses were 0, 10, 25 and 100 mg of glyphosate trimesium/kg body weight (bw)/day, given by gavage to maternal animals from the 7th day of gestation to postnatal day 11 (PND). The lowest observed adverse effect level (LOAEL) in the mother was > 100 mg, i.e. no maternal toxicity, considered harmful, was observed. In offspring, overall motor activity decreased (from 45 to 72 %) in males and females in groups exposed to 25 and 100 mg at the 14th day of pregnancy. Effects on learning and memory were also observed in offspring receiving the highest dose. These results were already recognised in the original 2001 study report, but the test laboratory considered them ancillary. By contrast, EPA recognised these effects and set a LOAEL for offspring at 25 mg/kg bw/day and a no observed adverse effect level (NOAEL) at 10 mg/kg bw/day. The EPA also considered the study acceptable for regulatory use. EFSA acknowledged these effects in discussions with experts from the Member States[186]. In the EU, NOAEL has been set at 50 mg/kg/day for glyphosate since 2017. The DNT study on Glyphosate-TMS indicates that NOAEL is in fact 10 mg/kg/day and | for adverse effects on reproduction and development. Regarding the neurotoxic effects of glyphosate-trimesium, please see replies to previous comments.<br><br># 140: No DNT study was included in the dossier. During the ongoing process for the renewal, EFSA was made aware that a guideline DNT study on glyphosate-trimesium was performed in 2001 (MRID 45539801); EFSA requested the applicants to make this study available. The study (using the trimesium salt) was submitted but not assessed by the RMS. The trimesium salt is considered a different substance than the acid form with evidence of neurotoxicity likely triggered by the trimesium ion (see US EPA assessment 2005). However, the study was assessed by the EFSA Working Group together with all the other available evidence available from the literature review on DNT potential of glyphosate (see Annex 7 of the Pesticides Peer Review Experts' Meeting TC 80). EFSA notes that no indication of neurotoxicity potential of glyphosate was present from the available data package for glyphosate (acid form).<br><br># 141: See reply to point # 140.<br><br># 142: Agree.<br><br># 143: During the Peer Review Expert TC 80 a data gap was identified to further investigate the cause of the observed effects from studies conducted with both GBHs and trimesium. As previously indicated, the paper by Pu et al., 2021 was not considered by the EFSA Working Group since being out of the time frame used for the literature review and not identified after the public consultation. The same applies to the study by Madani and Carpenter, 2022. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | therefore the EU reference values based on 50 mg/kg/day are not protective (applying a safety factor of 100, the ADI is currently set at 0.5 mg/kg instead of 0.1 mg/kg).<br><br># 139. On the other hand, recognising that the neurotoxic effects of Glyphosate-TMS on the developing foetus are relevant for the assessment of glyphosate could have led to a classification of glyphosate as presumed human reproductive toxicant category 1B in accordance with the CLP Regulation (EC) No 1272/2008. Moreover, Regulation 283/2013 requires the submission of developmental toxicity studies in order to determine reproductive toxicity, which demonstrates that the former is part of the second[187]. However, active substances falling within this classification cannot be approved under point 3.6.4 of Annex II to the PPP Regulation (except negligible exposure). Moreover, a classification for reproductive toxicity (including category 2) would have had the consequence that the metabolites of glyphosate would have been presumed to be relevant under the[188] water legislation.<br><br># 140. In any event, even if the applicants for re-approval were not required to provide the DNT study, it should at least have been quickly requested by the competent authorities (RMS or EFSA), considering that there is no other DNT study available, that Glyphosate-TMS causes adverse effects on offspring, and given its close structural similarity with glyphosate. According to the data requirements of Regulation 283/2013, " *If observations made in other studies or the mode of action of the test substance suggest it, additional* | Regarding Ojiro et al., 2023, please see reply to # 137 and the minutes of the Pesticides Peer Review Experts' TC 80. Regarding the assessment of studies by Costas Ferreira et al., 2022, von Ehrenstein et al., 2019 and Ongono et al., 2020, please see Annex 7 and Annex 4 of the Pesticides Peer Review Experts' Meeting TC 80.<br><br># 144: Differently from the case of chlorpyrifos and chlorpyrifos-methyl (where chlorpyrifos-methyl was considered a structural analogue of chlorpyrifos), glyphosate-trimesium salt is not considered a structural analogue of glyphosate and trimesium per se has a specific toxicological profile (US EPA 2015).<br><br># 145: See # 156-160. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | *studies or information may be required for the purpose of providing data on the postnatal manifestation of effects such as developmental neurotoxicity."*[189]<br><br># 141. In March 2022, Mie and Ruden (2023)[190] informed EFSA of the absence of the DNT study of Glyphosate-TMS in the re-approval application dossier. As indicated in the final RAR (Volume 1 p. 589), it was only at that time, in April 2022, that EFSA asked the re-approval applicants to make available the missing study. In its request, EFSA explains that the study can be considered relevant for the assessment of glyphosate, in particular in the absence of a DNT study for glyphosate acid and its other salts: "*It is acknowledged that the guideline DNT study was performed using the trimesium salt of glyphosate, whilst in the current application for renewal glyphosate acid and four other glyphosate salts are supported. Nevertheless, in the absence of DNT studies for glyphosate acid or any of its other salts, the existing DNT study on glyphosate trimesium may be regarded relevant in particular if there are scientific indications for potential harmful effects on human or animal health and should be considered by experts*" (emphasis added).<br><br># 142. In their reply, the applicants for re-approval explain that the DNT study on Glyphosate-TMS was not previously submitted because Glyphosate-TMS is considered to be an active substance different from glyphosate, with a distinct toxicological profile, and which has been withdrawn from the European market. This argument on the difference between the two | |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | substances is taken up by ECHA and by the RMS as explained in the final RAR.<br><br># 143. However, it must be recalled that it has not been demonstrated by the applicant or during the EU evaluation that Glyphosate-TMS salt should be considered as a separate active substance and not related to glyphosate acid or its other salts. This would require demonstrating that the adverse effects reported in the DNT study are due to trimesium ion and not to glyphosate itself. However, no other DNT studies were submitted or requested to demonstrate this. It should also be noted that, unlike the European authorities, the EPA included Glyphosate-TMS in its assessment of the carcinogenicity of glyphosate in 2017. Moreover, independent scientific literature clearly indicates that glyphosate (Pu *et al.*, 2021)[191], glyphosate isopropylamine salt (Ojiro *et al.*, 2023[192]) and various glyphosate products cause neurological effects in children or young animals (Costas Ferreira *et al. (2022)*[193], *Madani and* Carpenter, (2022)[194], von Ehrenstein e*t al.*, (2019)[195], Ongono *et al.*, (2020)[196]).<br><br># 144. It should be added that this rejection of the only DNT study carried out on a glyphosate salt contrasts strongly with the way in which, conversely, EFSA and Member State experts used chlorpyrifos data to conclude on the toxicity of a structural analogue, namely chlorpyrifos-methyl. The contrast is all the more striking because in the latter case a DNT study had indeed been carried out on chlorpyrifos-methyl, but had some flaws. | |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | # 145. The reason for these shortcomings is arguably to be found in the minutes of the EFSA Working Group. It states that ` *the RMS noted that RMS and MSs had limited time to review these data for discussion during the meeting*'[197]. In other words, due to the shortcomings in the re-approval dossier, the competent authorities did not have sufficient time to carry out a thorough developmental neurotoxicity assessment of glyphosate. However, a 'lack of time' cannot reasonably justify shortcomings in the assessment that would prevent a rigorous scientific risk assessment. | |
| 20. | Paragraphs 146 to 148<br><br>Unlawfulness and manifest errors in neurotoxicity assessment<br><br>(c) Insufficient regulatory studies | (c) Insufficient regulatory studies<br><br># 146. The results reported by the independent literature, together with the effects revealed by the DNT study on Glyphosate-TMS, point to serious and consistent concerns about the developmental neurotoxicity of glyphosate. Only particularly robust regulatory data would have been able to call those concerns into question. However, the renewal dossier contains only acute and sub-chronic toxicity studies, which are proven not to be adequate for the assessment of developmental neurotoxicity (Bloom and Boonstra, 2023[198]), in particular when this is manifested by behavioural effects. The same applies to neurodegenerative diseases, which occur at an advanced stage of life and, for that reason, are not covered by standard neurotoxicity tests. The *in vitro* tests (Tox21) that were submitted by the applicants for re-approval were not complete. As one Member State has pointed out, `*there is a remaining uncertainty because the assays included in Tox21 are not fully covering the current DNT in vitro test battery (Blum et al., 2023) and some processes were not tested*'[199]. | # 146-148. EFSA did not make use of information from acute and subchronic toxicity studies to conclude on the developmental neurotoxicity potential of glyphosate, but noted that the absence of signs of neurotoxicity in regulatory studies does not trigger the requirement to perform a developmental toxicity study as part of the regulatory dataset on the active substance. The availability of *additional data was however highlighted during the peer review and included an in vivo study in rats where DNT-related endpoints were assessed and considered as not affected by the high doses administered to dams, and ToxCast/Tox 21 data, where glyphosate was not showing any activity in all tested in vitro assays, except for one parameter at high concentrations. Further data, including public literature studies on glyphosate based herbicides and studies on other glyphosate salts (including glyphosate-trimesium), showing some DNT effects, were also assessed by the peer review.* No study was discarded and all the available information was integrated in a weight of evidence approach. EFSA concluded that there was no clear pattern of effects suggesting a DNT effect for *glyphosate,* and the current toxicological reference values were considered as sufficiently protective. However, EFSA |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | # 147. These shortcomings are not remedied by studies carried out in the context of the reproductive toxicity analysis, quite the contrary. The re-approval dossier does not contain an extended one-generation reproductive toxicity study (TG 443), even though such a study includes a set of developmental neurotoxicity parameters. Multi-generational studies have not tested any of these parameters with the exception of brain size. The most comprehensive of these studies is a two-generation toxicity study based on TG 416. The OECD itself acknowledges that, unlike the tests based on TG 443, '*OECD TG 416 lacks apical endpoints of developmental neurotoxicity, such as motor activity, sensory function, learning and memory*'[200].<br><br># 148. It follows from the foregoing that the serious indications of developmental neurotoxic effects from the scientific literature and the only available DNT study were discarded on the basis of regulatory studies which did not assess the essential parameters of developmental neurotoxicity. | still considered that there was a data gap related to the cause of the DNT effects seen in public literature studies with glyphosate-based herbicides (GBHs) and the study with *glyphosate-trimesium*. This data gap was reported as an outstanding issue in EFSA's Conclusion, but was not considered critical given the overall available data based on the weight-of-evidence.<br><br>EFSA made transparent reference to the weight-of-evidence approach and a data gap related to the cause of the DNT effects seen in the public literature studies with GBHs and the study with *glyphosate-trimesium* was reported as an outstanding issue. In so doing, it empowered risk managers to make a final decision, thereby considering the application of the precautionary principle. |
| 21. | Paragraphs 149 to 155<br><br>Unlawfulness and manifest errors in neurotoxicity assessment | # 149. At the end of their assessment, the experts of the EFSA led working group considered that the current set of data on glyphosate did not lead to the conclusion that there was no developmental neurotoxicity. More specifically, they acknowledged that in studies with alternative glyphosate salts or glyphosate-based formulations, there are indications of effects on DNT parameters and that in some studies these effects are accompanied by mechanistic data[201]. According to the minutes of the meeting, " *The EFSA working group considered that the potential of glyphosate active* | The *patere legem quam ipse fecisti* principle to which the requestor refers is better known in Union administrative law as the principle whereby, in the absence of legislative standards, and therefore in the presence of a considerable technical discretion, any Union Institution, body or agency is bound by its own determinations, and its discretion, where present, is limited by self imposed constraints. In this scenario, the competent Union administration is bound by its own policies or guidelines and must ensure not to infringe legitimate expectations that may have been created by these policies or guidelines, as well as |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | (d) A wrong and unlawful risk communication | *substance of affecting in vivo DNT endpoints cannot be concluded and that an additional uncertainty factor might be considered to cover this data gap*" (emphasis added). One Member State stressed that "*all data presented are indicative of a possible concern but how to address the DNT potential of GLY remains unresolved, and this is a data gap*"[202].<br><br># 150. As for EFSA, it acknowledged that the indications in the scientific literature are clear and that the current neurotoxicity studies submitted by the applicant have limitations to assess the DNT potential of glyphosate. It added that ' *it is however the full Body of Evidence (BoE), including mechanistic studies, which is raising the concern that based on this limited BoE, a DNT effect cannot be excluded. It is also important to consider that some endpoints are DNT specific and not really assessed in neurotoxicity studies (e.g. proliferation, migration, differentiation), therefore limiting the value of using neurotoxicity studies as a main, or exclusive, source of information*"[203].<br><br># 151. It appears from the content of these discussions that there is a real concern among Member States' experts about developmental neurotoxicity, and thus about the reproductive toxicity of glyphosate, which is a *cut-off criterion*. Therefore, the EFSA Conclusion should have referred to a "*critical area of concern*". Against all expectations, however, EFSA's conclusions merely express a "*lack of data" on the* subject, not a "*critical area of concern*". It states: " *Considering the overall body of evidence, a pattern of effects suggesting DNT liabilities was not clearly identified for glyphosate and the current toxicological reference values were* | compliance with the principle of equal treatment or non discrimination.<br><br>In the case at issue here, EFSA opted for limiting its technical discretion by defining the concept of critical areas of concerns as per the definition set out in the first paragraph of section 9.2 at page 32 of EFSA's conclusions. It is unclear, however, based on which arguments the requestor appears to conclude that EFSA exceeded the limits of the self imposed constraints set out in this definition by not identifying any Critical area of concern.<br><br>For what concerns the arguments raised by the requestor with regards the alleged flaws in terms of risk communication, EFSA would like to highlight that the conclusions of the peer review meeting referenced by the requestor as a matter of fact support the conclusions reached by EFSA.<br><br>Indeed, the reported conclusion of the peer review reflects the view of all experts but one, the opinion of whom has been transparently reported in accordance with the far reaching transparency standards characterising EFSA's operations. In this regard, ample evidence of EFSA's proactive and reactive transparency policies is available on its website, for instance at the following link: Transparency \| EFSA (europa.eu)<br><br>The data package on glyphosate (acid) did not indicate concerns triggering the requests of additional studies (namely a DNT study).<br><br>Extensive details on the scientific arguments behind the conclusion are reported in other sections of this document.<br><br>In addition, due attention should be paid that when assessing the complex scientific issues referred to it for |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | *considered protective. However, a data gap is identified for the applicants to clarify the cause of the DNT effects seen in the public literature studies with GBHs and in the study with glyphosate-trimesium*'[204].<br><br># 152. Even more, this '*data gap*' is included *in the list of 'other outstanding issues'* and not in the list of *'issues that could not be finalised*'. The distinction is far from negligible. The '*issues that could not be finalised*' are, in EFSA's terminology, related to data gaps which, once closed, may give rise to a concern or even a '*critical area of concern*' if this concern concerns all proposed uses of the representative formulation. With this formula, EFSA therefore draws the Commission's attention to the fact that the lack of data is particularly crucial in that it prevents the conclusion that there is no harmful effect on human or animal health and that there is no unacceptable effect on the environment. On the other hand, the '*other outstanding issues*' concern other data gaps which, although (allegedly) less '*critical*', must be considered relevant because of the uncertainties they pose to the assessment.<br><br># 153. In view of the above, the conclusion reached by EFSA is problematic and in breach of its own risk communication rules.<br><br># 154. On the one hand, it does not accurately reflect the content of the expert meeting's conclusions. The language used (some developmental neurotoxicity effects have not been clearly identified) is very different from the language used in those conclusions (there is a concern that a DNT effect cannot be excluded). | consideration, EFSA enjoys considerable discretion, which can be considered as exceeding it only insofar as it commits a manifest error of assessment. In particular, for what concerns paragraph 151 of the requestor's complaint, it remains unclear the reason why there should have been expectations that the data gap on reproductive toxicity results in a critical area of concern.<br><br>Furthermore, EFSA concluded it was appropriate to report under section 10 of its conclusions, amongst several other aspects, the need to obtain further clarification of the effects reported in the DNT study with glyphosate-trimesium and with other GBHs: indeed this information is relevant for Member States in the national process of authorisation of plant protection products. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | # 155. On the other hand, the finding of a mere '*lack of data*' and, more so, of a ' *pending issue*' is contrary to the definitions set out above and to which EFSA is bound by the *patere legem quam ipse fecisti* principle. As regards the lack of data, this qualifier seems manifestly inappropriate since, according to EFSA, there are no missing (regulatory) data and the results of the DNT study of Glyphosate-TMS do not call for the submission of a new DNT study. As regards the subsidiary classification of '*outstanding issues'*, it is even more incorrect since the 'question' concerns an exclusion criterion which concerns all representative uses, so that it was the qualifier of '*issues which could not be finalised*'[205] which, at the very least, should have been retained. | |
| 22. | Paragraphs 156 to 160<br><br>Unlawfulness and manifest errors in neurotoxicity assessment<br><br>Conclusion | # 156. It follows from the foregoing that the risk assessment of the neurotoxicity of glyphosate, including developmental neurotoxicity and thus reproductive toxicity, is vitiated by manifest errors of assessment and irregularities.<br><br># 157. First, many studies from independent scientific literature were not submitted by applicants for re-approval, in breach of Article 8 (5) of the PPP Regulation and Article 7 (1) (m) of Implementing Regulation No 844/2012. This mere fact should have led the RMS to conclude that the re-approval dossier was inadmissible. It was therefore not until the public consultation that some key studies were brought to the attention of the competent authorities. It is apparent from the minutes of the TC80 meetings that the experts did not have the time or even the opportunity to seriously address such studies, in breach of the principles of excellence and | #156-157. EFSA disagrees with the statement claiming to breach the principles of excellence and independence of the risk assessment.<br><br>A robust assessment of all available data has been undertaken in the context of the EU peer review. In addition to the RMS assessment, this included also an in-depth evaluation of the neurotoxicity data performed by the EFSA Working Group on glyphosate which was subsequently subject to an extensive scrutiny by MSs during the Pesticides Peer Review Experts' Meeting TC 80 before arriving to the final conclusions.<br><br>Indeed, as preparatory work to facilitate subsequent discussions in the expert meeting, the EFSA Working Group provided a thorough assessment of all the available information including appraisal of the published literature data, and a weight-of-evidence evaluation of the possible effects of glyphosate on human health as described in the |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | independence of risk assessment. Those studies were systematically declared unreliable or less relevant, with the result that regulatory studies were systematically given predominant weight, in breach of the *Blaise* judgment cited above (see *paragraph* 12). The lack of consideration for microbiome studies is, moreover, unlawful for the reasons set out above (see point 118 *above*).<br><br># 158. Secondly, a crucial regulatory study (on Glyphosate-TMS) was not submitted by the applicants for re-approval, in breach of several provisions of Implementing Regulation 283/2013 (points 1.1 to 1.3 of the introduction of its annex and point 5.6 of its annex). Neither EFSA nor the RMS (in breach of Article 11 (3) of Implementing Regulation No 844/2012[206]) showed due diligence since they did not identify this failure (even though contact with their US counterpart would have made it possible to identify it). When this study was finally submitted as part of the public consultation, its relevance was questioned on the grounds that Glyphosate-TMS was another active substance and did not have; the same toxicological profile as glyphosate even though EFSA initially considered this study relevant. This *ad hoc* statement of reasons is manifestly unfounded because (i) it contradicts a previous practice in relation to chlorpyrifos and chlorpyrifos-methyl (ii) the case-law allows read-across once the two substances at issue belong to the same group of chemicals and, overall, have a similar chemical structure; (iii) it leads to a reversal of the burden of proof since it presumes, against serious evidence from scientific literature, that the neurotoxic effects observed in the regulatory study on Glyphosate- | Annexes of the Peer Review Expert Meeting Report TC 80. This included also a rigorous assessment following a multistep and structured approach regarding neurotoxicity (cf Annex 7 of the Expert Meeting report TC 80).<br><br>Such extensive preparatory work and dedicated additional expertise provided by the EFSA Working Group prior to the expert meeting ensured solid basis for the discussions by the MS experts with a view to ultimately ensure robust conclusions to be drawn.<br><br>#158-160. The DNT study on trimesium was not submitted as part of the public consultation. The study was provided by the applicants following the specific request from EFSA in April 2022, once EFSA has been made aware of the availability of this guideline DNT study on glyphosate-trimesium.<br><br>Indeed no DNT study has been made available in the context of the EU evaluations on glyphosate, neither in the dossier submitted in the framework of the latest renewal process, nor in the previous renewal exercise.<br><br>EFSA acknowledged that the guideline DNT study was performed using the trimesium salt of glyphosate, whilst in the application for renewal glyphosate acid in the form of IPA salt was supported. Nevertheless, in the absence of DNT studies for glyphosate acid or its other salt(s), EFSA considered that such existing DNT study on glyphosate trimesium might be of potential relevance for both the renewal assessment of glyphosate, as well as for the classification and labelling process, and thus the applicant was requested to make this study available to both EFSA and ECHA as well as the RMS to allow consideration of all available evidence in the evaluations. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | TMS are caused by the trimesium salt and not from glyphosate.<br><br># 159. Third, the submitted regulatory studies contain limitations that do not allow for the assessment of certain aspects of developmental neurotoxicity highlighted in the independent literature and in the DNT study on Glyphosate-TMS.<br><br># 160. Fourth, EFSA's classification of this issue as a " *lack of data*" rather than a " *critical area of concern*" and, alternatively, as an "*open issue*" rather than as an "*issue that could not be finalised*" consists of a risk characterisation that (i) does not reflect the content of the discussions between experts and (ii) is not in line with its own rules. | EFSA confirms that no DNT study was available in the dossier, however it was considered not needed based on the lack of neurotoxicity effects in the regulatory dataset on glyphosate active substance. For this reason, no DNT study was highlighted as needed during the regulatory stop of the clock following the commenting phase.<br><br>During the risk assessment process, new evidence was brought forward, including public literature studies on glyphosate-based herbicides ('GBHs') in addition to the study on glyphosate-trimesium, showing some DNT effects. However, following a weight-of-evidence assessment, taking into account the overall body of evidence, EFSA concluded that there was no clear pattern of effects suggesting a DNT effect for *glyphosate* (see #146-148).<br><br>Nevertheless, to address the residual uncertainty, EFSA identified a data gap to clarify the cause of the DNT effects seen in the public literature studies with GBHs and in the study with glyphosate-trimesium. This conclusion was however not considered as critical in light of the overall body of evidence that also did permit the conclusion that the current toxicological reference values are considered as sufficiently protective. |
| 23. | Paragraph 161<br><br>Illegality and manifest errors in the assessment of endocrine disrupting properties | # 161. The assessment of endocrine disrupting properties is not based on a literature review published over the last 10 years. While the application for renewal was submitted on 13 December 2019, it appears from RAR[207] that systematic literature research was limited to studies published between 2014 and 2020. In other words, with the exception of a few studies taken into account in the previous re-approval procedure, studies published between 2010 and 2013 were neither comprehensively identified by applicants for re- | According to article Art 8(5) of the Regulation 1107/2009, *Scientific peer-reviewed open literature, as determined by the Authority, on the active substance and its relevant metabolites dealing with side-effects on health, the environment and non-target species and published within the last 10 years before the date of submission of the dossier shall be added by the applicant to the dossier.*<br><br>It is noted that although the application for renewal was submitted in 2019, the renewal dossier was submitted to |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | approval nor reviewed by the competent authorities. This omission, in breach of Article 8 (5) of the PPP Regulation and Article 7 (1)(m) of Implementing Regulation No 844/2012, sufficiently demonstrates that the assessment of the endocrine disrupting properties of glyphosate does not comply with the requirements for completeness or with the requirements of excellence intended to guide the risk assessment. | the RMS in June 2020. The literature search for all topics covered the period January 2010 - June 2020. In addition, specifically for the assessment of endocrine disruption, as stated in the RAR, as the previous literature search [33] covered the publication period between January 2014 and October 2016, a new search was conducted to cover the period from November 2016 to July 2019. Therefore, the literature search was in line with the legal requirement. It has to be further noted that additional literature papers considered in the previous assessments as well as literature brought to EFSA's attention during the public consultation and published after the commenting period (for the latter see excel file 'Consolidated list of newly available publications on glyphosate brought to the attention of EFSA and RMS after the public consultation phase until the time point of drafting the EFSA conclusion' [34] ) were also considered in the assessment for endocrine disrupting properties of glyphosate as well as for risk assessment. |
| 24. | Paragraphs 162-165<br><br>Illegality and manifest errors in the animal toxicity assessment<br><br>*Harmful effects on (farmed) animals* | # 162. According to Article 4 of the PPP Regulation, an active substance may be approved only in the *absence of "an immediate or delayed harmful effect on human* health (...*) or animal* health". Having regard to the definition of the environment given in Article 1(13) of the PPP Regulation, which includes *'wild* fauna', the concept of *'animal* health' must be understood as referring to animals which do not fall within the scope of *'wild* fauna'. It therefore covers, in particular, livestock[208]. | #162. Noted. EFSA does not have any specific comment on the point raised.<br><br>#163. The three studies by Ruuskanen *et al.* which are mentioned were assessed by the RMS and discussed during the Pesticides Peer Review Experts' Meeting TC 82. The peer review meeting agreed that not all of the assessed endpoints were relevant for assessing effects on the population. Moreover, as the tested formulated products were not the representative formulation under assessment, nor could their ecotoxicological comparability be confirmed, the studies were categorised 'less relevant but supplementary'. A data gap requesting that the applicant |

[33]undertaken in the context of a separate mandate (https://open.efsa.europa.eu/questions/EFSA-Q-2016-00663)
[34] Available in the Peer review Report in Open EFSA, Supporting documents section under EFSA-Q-2020-00140, refer to 'List of newly available publications (after commenting period)': Consolidated list of newly available publications after commenting period EFSA+AGG_March 2023_public.xls'

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | #163. In this respect, it should be noted that three studies by Ruuskanen *et al.* were discussed between experts from the Member States[209]. The authors exposed Japanese quails to doses of a glyphosate-based herbicide (Roundup Flex) representing 1220 % of NOAEL. Scientists have observed many effects: a disorder in the growth of feathers, an alteration of the intestinal microbiota, a drastic decline in testosterone, a reduction in the anti-oxidant activity of the liver, a change in embryonic development and an increase in brain oxidative stress of embryos. These 3 studies were considered reliable but discarded from the risk assessment (considered as *supplemental*) because the glyphosate-based herbicide used was not the representative formulation. However, in the absence of a regulatory study using the representative formulation, such studies should have been considered relevant. In addition, the magnitude of the impact of Roundup Flex on quails should have led to the RMS and EFSA asking applicants to repeat them using the representative formulation.<br><br># 164. The results of these studies have been corroborated in recent years by additional independent studies. Foldager *et al.* (2019)[210] assessed the impact of glyphosate concentration in industrial poultry feed and established a statistically significant negative correlation between the concentration of glyphosate in food and the hatching rate of breeding poultry eggs. In other words, the higher the concentration of glyphosate in food, the less eggs laid arrived at hatching. Estienne *et al.* (2022)[211], Freville *et al.* (2022)[212] and Estienne *et* | repeat the studies with the representative formulation was not concluded by EFSA as such studies are not required in accordance with Commission Regulations (EU) No 283/2013 and 284/2013. Furthermore, endpoints other than embryonic development and reproductive output were not considered relevant for the risk assessment which was performed in accordance with the agreed Guidance Document (EFSA, 2009[35]). The peer review concluded that the reliability of the endpoints should be reconsidered in line with the reliability criteria agreed during the meeting and, as a result, only some endpoints were assessed to be reliable by the RMS.<br><br>#164.<br><br>The studies of Estienne *et al.* (2022), Freville *et al.* (2022), Estienne *et al.* (2023) were published outside of the time period for which the assessment of the literature was performed.<br><br>The study of Foldager *et al.* (2019) is published in 2021 and therefore also falling outside of the of the time period of the literature search.<br><br>#165. As detailed above, the studies of Ruuskanen *et al.* were considered during the peer review. The studies of Estienne *et al.* (2022), Freville *et al.* (2022) and Estienne *et al.* (2023) were not considered as they were outside of the time period for which the literature search was performed. The study of Foldager *et al.* (2019) is published in 2021 and therefore also falling outside of the of the time period of the literature search. |

[35] EFSA (European Food Safety Authority), 2009. Guidance on Risk Assessment for Birds and Mammals on request from EFSA. EFSA Journal 2009;7(12):1438, 358 pp. doi:10.2903/j.efsa.2009.1438

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | *al.* (2023)[213] followed an identical protocol: 75 hens were exposed to a glyphosate-based herbicide (Gallup) at the dose of 47 mg/kg body weight, i.e. 47 % of NOAEL. The authors identified a series of statistically significant negative effects.<br><br># 165. It follows from these studies that no NOAEL can be set. Even at very low doses (12 mg/kg body weight in Ruuskanen studies), glyphosate-based herbicides have a harmful effect on the health of some farmed animals. In the absence of a regulatory study concerning the representative formulation, this finding should have been sufficient to conclude that the requirements of Article 4 (3) of the PPP Regulation were not complied with. In this respect, it should be added that the protocols used for the studies mentioned use animal cohorts significantly higher than those used in regulatory studies (e.g. 75 hens tested in Estienne 2022, Freville 2022 and Estienne 2023 compared to a maximum of 5 males and 5 females in regulatory studies). The statistical power of these studies is therefore significantly higher than that of regulatory studies. | Overall, reliability of the endpoints from the wild bird reproduction studies was discussed and agreed at the Pesticides Peer Review Experts' TC 82. The open literature studies discussed at the same meeting, were not considered to provide endpoints for the risk assessment.<br><br>Although the studies mentioned by PAN were not part of the peer review of glyphosate, it is noted that based on the scientific report from EFSA in 2018[36] on the evaluation of the impact of glyphosate and its residues in feed on animal health, the reported margin of exposure for poultry was 44[37], indicating a relevant margin of safety when compared with the agreed long-term endpoint. |
| 25. | Paragraphs 166-169<br><br>Unacceptable effects on insects | # 166. According to Article 4 (3) (e) an active substance may be approved only where it does not have an 'unacceptable effect on the environment', including wildlife. The toxicity study on insects other than bees is currently being conducted in accordance with the guidance document on terrestrial ecotoxicity (2002)[214]. This refers to ESCORT 2 for risk assessment. Escort 2 is | # 166. The risk assessment for non-target arthropods other than bees was performed following the tiered risk assessment scheme from the applicable guidance document (European Commission, 2002. Guidance Document on Terrestrial Ecotoxicology Under Council Directive 91/414/EEC. SANCO/10329/2002-rev. 2 final, 17 |

[36] https://efsa.onlinelibrary.wiley.com/doi/epdf/10.2903/j.efsa.2018.5283
[37] It is noted that the most up-to-date dietary burden value for poultry from the latest Art 10 MRL Reasoned Opinion (EFSA Journal 2021;19(10):6880), and considering all authorised uses, is lower than the one estimated in EFSA 2018.

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | the result of a workshop organised by Mr Candolfi (Novartis, now Syngenta), Chairman of the Organisation Committee and by employees of the pesticide industry (Novartis, Bayer, Zeneca, Huntington Life Sciences, JSC Int.), as well as an EU representative, a representative of the Dutch authorities and 2 representatives of the OECD. This document, written with a strong involvement of the pesticide industry, does not provide risk managers with informed conclusions on the toxicity of pesticides to insects. Indeed, it is based on a two-tier assessment. A first tier *using* two insect species considered to be 'most sensitive' in ESCORT2 (p. 8). If the toxicity of the pesticide tested exceeds a certain threshold, the applicant shall carry out additional toxicity tests with the two tier 1 species and add one or two other insect species of choice which are less sensitive to pesticides. The applicants consider that in the event that significant toxicity is established at tier 1, the RMS and EFSA should consider the pesticide to be excessively toxic to insects, instead of accepting testing with less sensitive species.<br><br># 167. The two tier 1 regulatory studies submitted by applicants for re-approval (dating from 1995) indicate very high toxicity of the representative formulation, namely 100 % mortality of insects (Aphidius rhopalosiphi[215] and Typhlodromus pyri[216]), at 'realistic' application rates, i.e. corresponding to authorised doses in some Member States. In the light of those results, those two studies alone should have been sufficient to | October 2002), which refers to the report from the ESCORT 2 workshop.<br><br>Regarding the regulatory studies,[38] the data set included (i) tier 1 studies with the standard species (*Aphidius rhopalosiphi* and *Typhlodromus pyri*) and the formulation for representative uses 'MON 52276' and (ii) extended laboratory studies (they can also be referred as tier 2 studies) with two standard species *A. rhopalosiphi* and *T. pyri*, as well as with the ground beetle *Poecilus cupreus*, the ground-dwelling spider *Pardosa* sp., the green lacewing *Chrysoperla carnea* and the rove beetle *Aleochara bilineata*.<br><br>All available studies and the risk assessment for non-target arthropods were discussed at the Pesticides Peer Review Experts' TC 82.<br><br># 167. Regarding the tier 1 studies, the experts at the meeting agreed that both studies with the standard species presented several limitations and, therefore, they were only considered as supporting information (i.e., the endpoints derived from the studies were considered unreliable and, therefore, they were not used to estimate hazard quotient (HQ) values for the in-field and off-field risk assessment).<br><br>The main reason questioning the reliability of the endpoint from the tier 1 (glass plate) studies with *A. rhopalosiphi* and *T. pyri* was that the test solution of formulated glyphosate used in the test produced a wet sticky layer on the treated glass plates that resulted in alterations of the moving behaviour of the arthropods to the point of sticking. Nevertheless, if the endpoints from the tier 1 studies would have been considered for the risk assessment and HQ values would have exceeded the trigger values, indicating |

[38] Relevant scientific peer-reviewed publications evaluating direct effects of glyphosate on non-target arthropods were not identified in the open literature in accordance with the criteria agreed at the Pesticides Peer Review Experts' TC 82.

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | conclude that there was no unacceptable risk to insects, especially since:<br>– They do not allow any LR50 to be set (see *above*, point 29).<br>– They concern two species of insect auxiliary to crops[217]. Destroying these insects therefore means depriving crops of natural biological control of certain pests, which *ultimately* leads to an increase in the need for the use of insecticides, contrary to the principle of integrated pest management which the Member States must implement under Article 14 of Directive 2009/128. The principle of sincere cooperation enshrined in Article 4 (3) of TEU obliges the Commission to assist the Member States in fulfilling their obligations under European Union law.<br><br># 168. However, ESCORT2 foresees additional studies to be carried out on both tier 1 species. However, it must be observed that no tier 2 studies were provided for Aphidius rhopalosiphi. In addition, other tier 2 studies show toxicity on insects at application doses similar to those authorised in the Member States. Indeed, an extended laboratory test study on *Typhlodromus pyri* (RAR Vol. 29, p. 493) indicates a negative impact on reproduction at a commonly used dose (6L/ha). Another study on *Chrysoperla carnea* (RAR Vol.29 p. 519) indicates a significant increase (+ 59 %) in offspring mortality at a dose close to the commonly used doses (12L/ha) and a significant reduction in the viability of laying eggs (-24 % at an application of 0,6L/ha). In other words, even if they were necessary – *quod non* – the tier 2 studies should | a high risk, the conclusions of the risk assessment would have not changed because, following the tiered approach in European Commission (2002) guidance (see above), the risk would have been refined with the available tier 2 (extended laboratory) studies with the and two additional ones.<br><br># 168. Regarding the extended laboratory studies, the experts also agreed that those with *C. carnea* and *A. bilineata* should be considered as supporting information.<br><br># 169. Therefore, the risk assessment for non-target arthropods other than bees was based on extended laboratory studies with the indicator species and with *Pardosa* sp. and *P. cupreus*. Since the rates causing 50% lethal and sub-lethal effects on all tested species were higher than the predicted environmental concentrations for the different representative uses with 'MON 52276', low in-field and off-field risk was concluded for such uses.<br><br>Overall, EFSA disagrees with the statement that risk assessment for non-target arthropods was not informative for risk managers. Indeed, according to the guidance currently in place, low in-field and off-field risk to non-arthropods other than bees was concluded for all representative uses. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | not have led to the conclusion that glyphosate had no unacceptable effect on insects. In addition, the maximum permitted doses in the re-approval regulation are similar to those leading to toxicity in regulatory tests: the Commission did not consider the results of the insect toxicity studies in its re-approval decision.<br><br># 169. However, in its Conclusion, EFSA does not provide any of this information to the risk managers. On the contrary, EFSA concludes that there is a *"low in- and off-field risk to non-target arthropods other than bees* (...) *for all the representative uses*" (p. 24 of the Conclusion). Such a conclusion does not, however, reflect the results of the studies provided by the applicants for re-approval. By failing to provide evidence of acute toxicity (100 % mortality for 2 insect auxiliary species in contact with the representative formulation, at a common application rate in the EU) or the impact of exposure to the representative formulation on the reproductive capacity of certain insects, EFSA failed to fulfil its obligations under precautionary principle[218], preventing risk managers from taking an informed decision. | |
| 26. | Paragraphs 170-172<br><br>Unacceptable effects on amphibians | # 170. The only regulatory test provided by applicants for re-approval for amphibians is a metamorphosis test on glyphosate alone. However, as explained above (see *paragraph 28*), this test demonstrates an effect of glyphosate on tadpoles at very low concentrations[219]. In particular, the distance from the snout to the vent is significantly increased in all concentrations tested, compared to the control. An increase in the incidence | # 170<br>The assessment of the endocrine disrupting properties of glyphosate was conducted following a structured and systematic approach in line with the ECHA/EFSA Guidance (2018) on the hazard identification of endocrine disruptors and thoroughly discussed with the EFSA Working Group on endocrine disruptors. All related documents are included in Annex 2 of the related background documents[39]. The |

[39] Available in the Peer review Report in Open EFSA, Supporting documents section under EFSA-Q-2020-00140, refer to the Peer Review Report: Part 3_Peer Review Report_Glyphosate_Annexes; Peer Review Report_Glyphosate_Annexes_TC84_16 August 2023_EFSA: Annex 2. EFSA ED WG Advice Non-target organisms (NTOs)

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | Lack of excellence and independence of scientific assessment / underweighting independent literature data | of mild thyroid hypertrophy and an increase in follicular cell hypertrophy shall be reported. However, those effects were discarded by the RMS and EFSA, which concluded that those effects were not related to a disturbance of the thyroid system, without any scientific justification (p. 63, RAR19).<br><br># 171. It should also be noted that this test concerns only the development of tadpoles. However, juvenile and adult amphibians live in agricultural areas and are likely to be exposed by direct spraying in the field or on the edge of the field. For example, Relyea (2005)[220] indicates that exposure of tadpoles or frogs to Roundup Weed and Grass killer at a concentration of 3.8 mg/L glyphosate causes mortality of 100 % of tadpoles and at least 68 % of juveniles depending on the species tested.<br><br># 172. Many other studies highlight the toxicity of glyphosate on amphibians and in particular frogs[221]. However, this independent literature is systematically considered irrelevant because it does not relate to the representative formulation. However, this systematic exclusion is not in line with the principle of excellence in scientific evaluation. In the absence of comparable studies on the representative formulation, such studies should have been considered relevant for determining the effect of glyphosate "*under realistic conditions of use*"[222], i.e. in plant protection products. | assessment was extensively discussed at the Pesticides Peer Review Experts' Meeting TC 84.<br><br>EFSA disagrees with the statement related to the available Amphibian Metamorphosis Assay (AMA), referred to in column 2. Indeed, this study was overall considered negative since it did not show a pattern of endocrine activity and/or adversity. In the study, no change in the developmental stage and/or nHLL (normalised Hind Limb Length) was observed in tadpoles exposed to 5 concentrations of glyphosate active substance. Only a very slight increase in the prevalence of thyroid gland hypertrophy and follicular size increase was observed at the highest tested concentration. However, this was only mild and not observed with a dose response. The effects in snout–vent length (SVL) were observed at the 3 highest tested concentrations. However, there was no dose response.<br>It has to be considered that while developmental stage and nHLL are T-mediated parameters, SVL is a 'sensitive to, but not diagnostic' parameter, according to the classification in the ECHA/EFSA Guidance on the hazard identification of endocrine disruptors.<br>As outlined above, a conclusion that glyphosate does not meet the criteria according to point 3.8.2 of Annex II to Regulation (EC) No 1107/2009, as amended by Commission Regulation (EU) 2018/605, was reached following a weight of evidence approach in line with the ECHA/EFSA Guidance (2018) on the hazard identification of endocrine disruptors.<br><br>#171. Several scientific peer-reviewed open literature studies were available which investigated the effects of glyphosate formulations on terrestrial phases of |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | | amphibians. The criteria for assessing the relevance and reliability of the studies were discussed and agreed at the experts' meeting (Pesticides Peer Review Experts' TC 82). Few studies were considered to provide endpoints which are potentially relevant to populations. However, when considering the available information, adverse and biologically relevant endpoints were not obtained.<br>Relyea (2005) was not explicitly considered in the latest analysis of the literature, as it was outside of the temporal range and it did not report relevant data (a formulation containing POEA was tested).<br><br># 172. All endpoints currently listed in the List of Endpoints attached to the EFSA conclusion are in fact proceeding from open literature studies. Based on these data, a comparison of the hazard data with fish was carried out and discussed at the Pesticides Peer Review Experts' TC 82.<br>For acute (lethal) effects due to exposure to glyphosate, the lowest fish endpoint was agreed to be protective for amphibians. For chronic exposure to glyphosate, a proper comparison between fish and amphibians could not be carried out, since relevant and reliable chronic endpoints for amphibians were not available. A full comparability between fish and aquatic stages of amphibians would anyway be hampered by the different response types being measured for the two groups. |
| 27. | Paragraphs 173-176<br><br>Manifest errors of assessment on effects on environment - | # 173. The assessment of the toxicity of glyphosate to animals is not in line with the standard of excellence of the risk assessment.<br><br># 174. First, it does not support the conclusion that there is no harmful effect on animal health. Indeed, studies from independent literature indicate a | # 174. see reply to #165<br><br># 175. As explained in paragraphs #166-169, the risk assessment for non-target arthropods was carried out according to the stepwise approach included in the guidance document currently in place. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | Conclusion | significant effect on the breeding of poultry at extremely low doses. Such studies could not reasonably be disregarded either on the ground that they do not concern the representative formulation – although similar studies concerning that formulation are lacking – or on the pretext that they would be contradicted by regulatory studies relating to the active substance alone – even though they are carried out on a much smaller number of animals.<br><br># 175. On the other hand, that assessment also does not support the conclusion that there is no unacceptable effect on the environment. The two tier 1 regulatory insect studies showed a mortality rate of 100 % in crop auxiliaries. One of these studies was not followed by a tier 2 test, which was required. Other tier 2 tests revealed significant adverse effects. As for the effects on amphibians, the only regulatory study carried out showed an adverse effect on tadpoles but was rejected by the competent authorities without proper justification. In addition, independent literature documents significant toxicity of glyphosate-based herbicides. Again, that literature could not be disregarded simply because it does not concern the representative formulation since a regulatory study to test the toxicity of that representative formulation on amphibians was not carried out by the applicants for re-approval.<br><br># 176. Therefore, by discarding or underweighting independent literature data as well as positive results | For amphibians, the regulatory study (i.e. the AMA test), as explained in the response to #170, did not show a pattern of endocrine activity and/or adversity.<br><br>It is noted that there are no specific data requirements for regulatory studies on amphibian and reptiles and for a specific risk assessment, but literature data were provided and considered according to the data requirement in point 8.1.4 of Commission Regulation (EU) No 283/2013.<br>Criteria for evaluation of the literature data for their relevance and reliability were extensively discussed and agreed with the experts (see expert consultation point 5.10 of the meeting report of the Pesticides Peer Review Experts' TC 82[40]) and subsequently the studies were reconsidered by the RMS according to those criteria. Therefore, EFSA disagrees with the statement in # 176. |

[40] available in the Peer Review Report in the Open EFSA under 'Supporting documents' under EFSA Question number EFSA-Q-2020-00140: https://open.efsa.europa.eu/study-inventory/EFSA-Q-2020-00140 ; refer to Part 3_Peer Review Report_Glyphosate_expert meeting reports_public.pdf (TC 82, Expert consultation point 5.10)

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | from regulatory testing, by confining themselves to incomplete regulatory data, and by failing to adequately report on the reported risks, the competent authorities committed manifest errors of assessment and failed to comply with the evaluation requirements. | |
| 28. | Paragraphs 181-185<br><br>Illegality and manifest errors in the assessment of the effect of glyphosate on biodiversity<br><br>Poorly listed independent scientific literature | # 181. There is a wealth of independent scientific literature documenting the biodiversity effects of the use of glyphosate products. By way of example, Boutin *et al.* (2014)[223] points to the reduction in the production of seeds from wild plants because glyphosate is often sprayed at the plant breeding stage. Damsgaard *et al.* (2014)[224] report wild plants (perennial species) in semi-natural plant communities that are significantly affected by glyphosate by spraying drift. *Strandberg et al.* (2012 and 2021)[225] criticize the current risk assessment of pesticides, which was considered not to be sensitive, and concludes that the levels of glyphosate representative of the spray drift have led to a loss of biodiversity and a change in species composition (on the edge of the fields). Baker *et al*. (2014)[226] report indirect effects of glyphosate. They show that due to the reduction of macrophytes due to exposure to glyphosate in wetlands, the abundance of chironomids has increased. Another study by Baker *et al.* (2016)[227] demonstrates variations and indirect effects on zooplankton and phytoplankton communities during exposure to realistic levels of glyphosate. Furthermore, although there is no authorisation to use glyphosate in wetlands, the article de Silva *et al.* (2023)[228] established | A summary of the biodiversity assessment is reported in the RAR, Volume 3 – B.9 (PPP), B.9.14.1[41]. *Assessment of risk to biodiversity via indirect effects and trophic interactions*.<br><br>The assessment was extensively discussed at the Pesticides Peer Review Experts' Meeting TC 82 (expert consultation point 5.25[42]).<br>The experts recognised that literature search according to the principle of the systematic literature review was not available and a data gap was identified by EFSA in section 10 of the EFSA Conclusion (2023) (i.e. *to perform a systematic literature search for data collection)*.<br><br>Criteria for evaluating the papers for their relevance and reliability were also discussed at the TC 82. The available papers were then re-evaluated by the RMS in line with the agreed criteria, as reported in Appendix A to RAR Volume 3 – B.9 (PPP) MON 52276 Literature data on biodiversity.<br><br>It is noted that the papers mentioned in #181 and #182, i.e. Boutin *et al.* (2014), Damgaard *et al.* (2014), *Strandberg et al.* (2012 and 2021), Baker *et al*. (2014), Baker *et al.* (2016), Mudge and Houlahan (2019), Van |

[41] available in the Peer Review Report in the Open EFSA under 'Supporting documents' under EFSA Question number EFSA-Q-2020-00140: https://open.efsa.europa.eu/study-inventory/EFSA-Q-2020-00140 (refer to Glyphosate_Final RAR_public.zip)

[42] available in the Peer Review Report in the Open EFSA under 'Supporting documents' under EFSA Question number EFSA-Q-2020-00140: https://open.efsa.europa.eu/study-inventory/EFSA-Q-2020-00140 (refer to Part 3_Peer Review Report_Glyphosate_expert meeting reports_public.pdf (TC 82, Experts' consultation point 5.25)

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | that it is detected in almost all environmental matrices, so it is important to assess its effects also beyond the spray fields. Mudge and Houlahan (2019)[229] also showed a decrease in the richness of macrophytes species, an increase in the macrophyte species evenness, a decrease in macrophyte coverage and a reduction in the similarity of communities during exposure to glyphosate.<br><br># 182. While the above mentioned articles were discussed by the RMS and/or experts (TC80), many others were not discussed. De Lima Silva & Pelosi (2023)[230], for example, concludes that if the tests required by Implementing Regulation 283/2013 are insufficient to reveal effects on earthworm populations, these effects are clear when testing includes more parameters: `*under more realistic conditions, that is, when assessing sensitive endpoints (e.g., reproduction, growth) and using species present in the field, after several applications per year, the negative effects of glyphosate or GBH on earthworms were observed at the subindividual, individual, population, and community levels, as well as on earthworm-mediated functions*'. Sanchez-Bayo (2021)[231] notes that "*residues of herbicides can reduce the biomass of macrophytes in ponds and wetlands, indirectly affecting the protection and breeding of predatory insects in that environment*". Van Bruggen *et al*. (2021)[232] report another indirect effect, namely the alteration of microbial communities as a result of exposure to glyphosate: *Plant growth promoting rhizobacteria and beneficial intestinal bacteria often are negatively affected, while pathogenic bacteria and fungi are enhanced*. Vera *et al*. (2012)[233] report direct and indirect effects on microbial | Bruggen *et al*. (2021), Newman *et al*. (2016), Motta *et al*. (2018), were considered during the peer review.<br><br>Vera *et al*. (2012) is not available in the RAR since it was excluded based on title and abstract.<br><br>The papers from Silva *et al.* (2023), De Lima Silva & Pelosi (2023) were not peer reviewed since they were published after the publication of the EFSA Conclusion. Sanchez-Bayo (2021) and Ruuskanen *et al.* (2023) were not considered in the peer review, since they are outside the time period of the literature search and, it is further noted, they are not specific for glyphosate.<br><br>EFSA disagrees with the statement in #183 that the references were not examined, since, although the literature search was considered lacking an appropriate problem formulation, search strategy and methodology, all the papers brought to the EFSA attention during the peer review were evaluated or screened.<br><br>#184-185: Noted. EFSA has no additional comment. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | freshwater communities. Newman *et al*. (2016)[234] report changes in soil communities: "*in the presence or absence of glyphosate, corn and soybean rhizospheres were dominated by members of the phyla Proteobacteria, Acidobacteria, and Actinobacteria. Proteobacteria (particularly gammaproteobacteria) increased in relative abundance for both crops following glyphosate exposure, and the relative abundance of Acidobacteria decreed in response to glyphosate exposure*.' Motta *et al*. (2018)[235] show that glyphosate can disrupt honeybees microbiome and thus indirectly bee health. Ruuskanen *et al.* (2023)[236] conclude that '*herbicides can influence natural and agricultural ecosystem functioning due to soil- and host-associated microbiome alteration and may have evolutionary consequences*'. They also conclude that impacts on biodiversity and ecosystems cannot be studied in the laboratory and that field studies are needed.<br><br># 183. The reason why these (and other) references were not examined is due to the very low quality of the state of the literature carried out by the applicants for re-approval. The experts themselves regretted the fact that the criteria used to identify the articles were unclear[237]. In particular, the experts considered that even studies concerning the use of a different formulation than the representative formulation were potentially relevant[238].<br><br># 184. More severely, it appears that:<br>– The applicants were requested to provide a revised biodiversity assessment including new data collected in a systematic manner, however no new study were provided by the applicants; | |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | – Part of the studies presented in the biodiversity assessment were retrieved by the RMS from the general literature search;<br>– During the peer review the applicants were requested to justify the absence of certain relevant studies from the results of the literature search, but no justification was provided.<br>The experts agreed with the concern of the RMS regarding the lack of an unbiased or systemic approach to the data collection. The experts also suggested that the applicant's approach was lacking a quantification of the level of effect via indirect effects on biodiversity.[239]<br><br># 185. In the light of these factors, there is no doubt that the applicants for re-approval failed to fulfil their obligations under Article 8 (5) of the PPP Regulation and Article 7 (1) (m) of Implementing Regulation No 844/2012. For this reason, the rapporteur Member States should have declared the dossier inadmissible under Article 8 (6) of Regulation No 844/2012. These deficiencies prevented a full and fully informed assessment of the results of the scientific literature by the RMS and, in turn, by EFSA and then by the Commission. | |
| 29. | Paragraphs 186-191<br><br>Illegality and manifest errors in the assessment of the effect of | # 186. These deficiencies in the assessment of independent literature are aggravated by the fact that no field study to assess the impact of glyphosate on biodiversity has been provided by the applicants for re-approval.<br><br># 187. Instead, they provided a study entitled ` *Glyphosate: Indirect effects via Trophic interaction – A* | In response to #186, it is noted that there are no specific data requirements, evaluation/decision making criteria and harmonised approach for assessing biodiversity and indirect effects. Therefore, the lack of field study cannot be considered as non-fulfillment of the legal requirements; furthermore, it is questionable whether one study could have been sufficient, by considering that, for example, in the report from Kemi (Swedish Chemicals Agency) |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | glyphosate on biodiversity | *Practical Approach to Biodiversity Assessment*'[240]. Although the content of this study is confidential, it seems that it proposes to use the category of *'specific protection goals'* already existing in certain EFSA documents.<br><br># 188. The approach followed in this study was considered ` *inadequate*' and ` *of questionable scientific quality*' by experts[241]. Furthermore, it appeared that that study did not apply the methodology proposed to the use of glyphosate and therefore did not propose any conclusion as to the acceptability of the environmental risk posed by this substance when indirect effects, including trophic interactions are taken into account. Indeed, the study concludes that:<br>> the risk to diversity and abundance of non-target arthropods and vertebrates via trophic interactions will depend on the specific agricultural and local ecological conditions and that mitigation measures need to be adapted to the landscapes surrounding fields in the respective MS[242].<br><br># 189. It is remarkable to note from that conclusion that the study is based on the assumption that the risks posed can necessarily be maintained at an acceptable level by risk reduction measures. This is a fundamental misunderstanding of the PPP Regulation, which implies, on the contrary, from the stage of approval of the active substance, the demonstration of the acceptability of the risk, at least for certain representative uses of a formulation, under realistic conditions of use. This acceptability must of course be demonstrated by taking into account, cumulatively, direct (ecotoxicology) and | mentioned in column 2, multiple approaches would be needed to address the issue. It is also noted that it is generally final applicant's responsibility to decide on the strategy for addressing particular issues such as the provision reported in Annex 1 of Commission Implementing Regulation (EU) 2017/2324 for MSs to pay particular attention to *the risk to diversity and abundance of non-target terrestrial arthropods and vertebrates via trophic interactions.*<br><br>Regarding #187, applicant submitted a report which is summarised in the RAR, Volume 3 – B.9 (PPP), B.9.14.1.<br>In response to #188, the report was extensively considered during the peer review and the outcome was a data gap, as reported in section 10 of the EFSA Conclusion, in consideration of the lack of a harmonised approach and specific protection goals (SPGs).<br><br>In response to # 189, it is noted that, as part of the data gap, EFSA reflected the need to consider the effectiveness of possible risk mitigation measures at landscape level, for all the uses being assessed.<br><br>#190. Noted. The statement reflects the text of data gap written in the EFSA (2023) conclusion.<br><br>Regarding comment in #191, EFSA appreciated the effort of the German authority Umwelt Bundesamt (UBA) to define a method that could help MSs to cover indirect effects. UBA's proposal is grounded on a screening approach that, in their opinion, should be potentially useful as interim solution for the authorisation of the PPPs. However, EFSA notes that the development of an intermediate or provisional methodology was not included |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | indirect (biodiversity) effects on the environment, and not through an isolated approach to both dimensions. At the very least, it is surprising that applicants for re-approval did not even try to demonstrate this.<br><br># 190. However, according to the experts' opinion, it was possible to identify a method capable of assessing the impact on biodiversity. According to them, '*It was reflected that the lack of harmonised approaches to the assessment of biodiversity poses challenges. Nevertheless, a sound assessment could have been performed by the applicants. These were reflected in the document that was produced by the EFSA Working Group (WG) (Annex to this document)*'[243]. A meeting took place on this issue between the RMS, the applicants and EFSA: *"It is important to point out that the general approach used for the biodiversity assessment was discussed with the RMS and their feedback was then used to develop the final approach for the assessment. EFSA was an observer to this meeting and offered generic approaches that involved developing landscape level models*'[244]. Moreover, EFSA's opinion states also the way forward for such an assessment:<br>For further addressing the risk to biodiversity via indirect effects and trophic interactions it was considered needed (1) to perform a systematic literature search for data collection; (2) to quantify, in a spatial and temporal context, the direct effects on the weeds (including the impact on the seed bank), non-target plants, non-target arthropods and bees in order to inform the extent of potential indirect effects via trophic interactions; (3) to | for consideration amongst the prioritisation exercise carried out by the European Commission, EFSA and MSs, to which Germany also contributed in the context of the SCoPAFF discussions. Therefore, it remains unknown to EFSA whether an interim solution is also envisaged or supported by other MSs.<br>Regarding the report from Kemi (Swedish Chemicals Agency), EFSA would like to note that as matter of fact, contrary to what the complainant states, no specific method is actually proposed in this report. However, it includes a systematic literature review of existing approaches, and the authors concluded that a combination of those different approaches would be needed to address the evaluation of biodiversity and indirect effects e.g. semi-field, field studies, modelling along with monitoring would be useful. These recommendations are valuable and can be considered in the context of the discussion within next developments for a harmonised method.<br><br>No agreement on the methods to be used is yet available. Considering the complexity of the topic, a robust approach is deemed necessary to ensure that indirect effects are properly introduced and implemented. To this purpose EFSA considers as priority the definition and agreement of SPGs for non-target arthropods and non-target terrestrial plants. Indeed, arthropods and wild plants are fundamental entities of food networks and preserving both their biomass and diversity is pivotal to safeguard ecosystem services delivery and ecological function such as habitat provision, food web support, pest control, pollination. It is therefore clear that the updating of the guidance documents on non-target arthropods and non-target terrestrial plants is of fundamental importance also for the scientific evaluation of indirect effects. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | demonstrate how both specific and general mitigation measures may address the impact due to indirect effec[245].<br><br># 191. Methods for assessing the impacts of pesticide use on biodiversity have also been developed at national level for the authorisation of plant protection products, for example in Germany[246] or Sweden[247]. It would therefore have been easy to use one of these methods pending the adoption of harmonised guidelines at European level. | |
| 30. | Paragraphs 192-199<br><br>Biodiversity - No risk assessment in absence of guidance document<br><br>Erroneous conclusion | # 192. The Member States and EFSA reported that there was no guidance document on assessing the impact of active substances on biodiversity and concluded that there was a lack of data.<br><br># 193. This conclusion is erroneous.<br><br># 194. First, it does not reflect the content of the discussion between experts from the Member States, who, on the contrary, concluded that '*a sound assessment could have been performed by the applicants*', on which it seems that at least one meeting took place with the renewal applicants.<br><br># 195. Secondly, the PPP Regulation does not provide that the consideration of biodiversity effects is subject to the adoption of a guidance document. At most, Article 4 (3) (e) provides that such an assessment must take place '*when the scientific assessment methods of these effects, accepted by the Authority, are* available'. Article 4 therefore allows EFSA to accept these assessment methods on *an ad* hoc basis as part of a | # 192. Noted. EFSA has no additional comment.<br><br>EFSA disagrees with the statement in paragraph #193; indeed the lack of a harmonised approach to assess biodiversity, as it was acknowledged during the peer review of glyphosate, is factual. Despite some scientific insights and developments in the area, a validated approach agreed by the European Commission and MSs is not yet available.<br><br>The '*sound assessment*' refered to in paragraph #194 is explained in the position paper of the EFSA Working Group (WG), where some recommendations were given to address the issue in the absence of a harmonised approach. These recomendations are very specifc and do not refer to any existing approach in particular.<br><br>On the contrary, there was a general agreement that the issue could be addressed during the development and agreement on specific protection goals for non-target organisms. Indeed, SPGs allows the implementation of the generic protection goal of Article 4(3)(e) of Regulation (EC) No 1107/2009 to have "no unacceptable effects on the |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | dossier. This is, moreover, what EFSA and the RMS tried to do in this dossier, in vain. In any event, under Article 4 (3) (b), the taking into account of indirect effects on animal health is not conditional on the adoption of such methods.<br><br># 196. Third, it is implausible to conclude that "there is a *lack of data* " in view of the wealth of studies from independent literature. Rather, the problem lies in the fact that this literature was not systematically collated by applicants for re-approval. For example, on aquatic organisms, experts find that '*No conclusion can be reached given the lack of systematic literature search*'[248].<br><br>*Conclusion*<br><br># 197. It follows from the above that the biodiversity risk assessment does not meet the requirements of completeness and excellence.<br><br># 198. On the one hand, as the experts have repeatedly pointed out, independent scientific literature has not been properly identified, summarised and communicated by the applicants, so that it could not be properly reviewed. This alone should have led the RMS to declare the renewal dossier inadmissible. The additional requests for information and the proactive research carried out by the RMS have only permitted to fill these gaps in part. | environment", through a structured and transparent definition of what to protect, where to protect it, to which extent, over what time period and with which degree of certainty.<br><br>Therefore, EFSA disagrees with the statement in paragraph #195, because it is clear that specific methods accepted by the Authority are not available, despite the effort made by EFSA during the peer review of glyphosate to guide the applicants by way of a formal request to empower them to submit a scientific assessment compliant with the recommendations as set out in the data requirement issued for the applicants[43]. Since a harmonised approach requires the definition of SPGs and/or specific data requirements, the adoption and agreement of such approach, once available, is fundamental.<br>In response to # 196, as reported in the EFSA Conclusion, most of the studies were considered to be of low relevance for the representative uses, therefore the experts agreed that a conclusion cannot be reached to exclude possible negative impacts on non-target species, habitats and ecosystems due to indirect effects via trophic interactions.<br><br>In response to # 197 and # 198, a data gap was identified in section 10 of the EFSA Conclusion.<br><br>In response to #199, according to Art 13 of Regulation (EU) No 844/2012, *the Authority shall adopt a conclusion in the light of current scientific and technical knowledge using* guidance documents available at the date of the submission of the supplementary dossiers...'. |

[43] See Reporting Table point 5(435) available in the Peer Review Report in the Open EFSA under 'Supporting documents' under EFSA Question number EFSA-Q-2020-00140: https://open.efsa.europa.eu/study-inventory/EFSA-Q-2020-00140 (refer to Part 2_Peer Review Report_Glyphosate_reporting tables_public.pdf (electronic pages 1870-1872 of 2930)

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | # 199. On the other hand, the applicants did not even attempt to demonstrate that the use of glyphosate-based formulations was not likely to have unacceptable effects on the environment, when they include effects on biodiversity (indirect effects in particular via trophic interactions). Similarly, the competent authorities have confined themselves to noting a lack of data, while the real lack is a general methodology for assessing the risks to biodiversity. However, methods exist, notably at Member State level, so that it was open to the authorities to propose them on an *ad hoc* basis, in the absence of a proper guidance document which was awaited since the entry into force of the PPP Regulation 15 years ago. | It is noted that the Kemi (Swedish Chemicals Agency) report was published in 2021, and the approach developed by UBA (German Environment Agency – Umweltbundesamt) was provided to EFSA in 2022, therefore, they were not yet available at the time when the applicants submitted their dossier (June 2020).<br>Furthermore, 4(3)(e)(iii) of the Regulation 1107/2009 prescribes that no "unacceptable effects" on the environment, shall be identified. The concept of "unacceptable effects" is in itself rather generic and implies quality judgements which pertain to the sphere of responsibility of the risk managers. In the absence of the setting of specific protection goals clarifying the concept of what is deemed unacceptable, EFSA is in turn not in a position to identify "accepted scientific methods" to assess indirect effects.<br>No provision under the Uniform Principles explicitly empowers EFSA to adopt binding guidance for the definition of criteria or standards to be applied in the context of its scientific evaluations, or in the context of the scientific evaluations of the Rapporteur Member States.<br>Therefore, it is clear that EFSA may not proactively implement guidance documents and must react to a mandate received from the Commission to develop any guidance and to the endorsement at the SCoPAFF to apply it.<br>Overall, on the basis of the above background, it is EFSA's opinion that the implementation of any 'interim solution' without a large consensus and agreement from European Commission and MSs, is therefore not an option that could have been considered. |
| 31. | Paragraphs 200 - 201 | # 200. A number of publications highlight that glyphosate is ubiquitous in the air in the EU. For | EFSA does not consider the performance of a long-term inhalation toxicity study as a relevant study requirement for |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | Failure to take into account exposure to glyphosate by inhalation | example, glyphosate was found in the air in 100 % of the 69 sites tested across Germany (Kruse-Plass *et al.,* 2021[249]). The study also showed that glyphosate is transported over long distances, to the heart of nature reserves or forests. A similar study in Austria (Zaller *et al.,* 2022[250]) identified glyphosate in the air of the 15 sites tested. In 2024, the NGO Generation Futures published a report concluding that glyphosate is, qualitatively and quantitatively, the most found pesticide in the air in northern France. Furthermore, glyphosate is frequently found in house dust, as stated in a recent publication (SPRINT, Navarro *et al.,* 2023[251]): of the 190 pesticide substances found, glyphosate is the one with the highest median concentration and the 2nd pesticide most often found in farmers' homes, while it is the pesticide most present in dust in private homes, together with dozens of other pesticides. Citizens are therefore, for some, continuously exposed via inhalation.<br><br># 201. In view of that finding, it must be observed that the inhalation risk assessment carried out by the RMS and EFSA does not support the conclusion that there is no harmful effect on health. Indeed, according to the Guidance Document[252], this assessment is limited to the effects related to inhalation exposure of 24 hours. In other words, no long-term assessment has taken place despite reading independent scientific literature, the majority of the European population is permanently exposed to glyphosate by inhalation. Since exposure to glyphosate via air is a long established fact (Chang *et al.*, 2011[253]), the RMS and EFSA should therefore have | glyphosate. No local effects in the respiratory tract are anticipated at the expected concentrations of glyphosate in the air compartment or in house dust. Those levels would likely contribute in a marginal way to the overall systemic exposure to glyphosate for the general population**.** From the review of available biomonitoring data performed in the peer review, estimated systemic exposure levels (resulting from different exposure pathways in the population, including by inhalation) were below the derived toxicological reference values for the EU population.<br><br>Regarding the monitoring data of glyphosate in the air compartment, despite the few data available and the intrinsic properties of glyphosate (i.e. non-volatile), the information provided from literature review (including Kruse-Plass et al., 2021) showed a high frequency of quantified samples with values >LOD (limit of detection) for glyphosate. However, the sampling apparatus (passive samplers) used in these studies measured particulate-bound glyphosate and not gas phase only. Transportation to air was therefore likely to be caused by wind-eroded particles transportation rather than volatilisation or transport of aerosols formed during spraying.<br><br>Regarding human health (as the Uniform Principles indicate that air concentrations are to be assessed against the human-toxicological threshold), the assessment for bystanders/residents was carried out with the EFSA model (belonging to the EFSA operator exposure guidance[44]). The default concentration in air was 1 microgram/$m^3$ (hence 0.001 mg/$m^3$), resulting in systemic exposure estimations for residents and bystanders below the (A)AOEL for all the representative uses proposed in the RAR. The risk |

[44] EFSA (European Food Safety Authority), 2014. Guidance on the assessment of exposure of operators, workers, residents and bystanders in risk assessment for plant protection products. EFSA Journal 2014;12(10):3874, 55pp.https://doi.org/10.2903/j.efsa.2014.3874

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | asked applicants for a long-term inhalation toxicity study (aerosols, dust particles loaded with glyphosate), in particular on the basis of the environmental concentrations available in the scientific literature. In the absence of such a study, EFSA could not exclude a high risk to health, in particular that of residents and in particular that of young children. | characterisation is presented in the toxicology section. This default was above the monitored concentrations determined in air samples.<br><br>The cited open literature in # 200 Zaller *et al.,* 2022 and SPRINT, Navarro et al., 2023 were not considered in the peer review evaluation. Formally, in line with the legislation, there is no legal obligation to consider newly available data submitted outside of the dedicated public and targeted consultations or after the deadline of the window for providing the additional information within the clock stop period, unless they constitute adverse data (cf Article 56 of Regulation (EC) No 1107/2009 regarding information on potentially harmful or unacceptable effects).<br><br>Indeed the quoted studies were published after the timeframe of the literature search performed according to the regulatory requirement and they were also not brought to EFSA's attention following that period. |
| 32. | 201/1.10<br><br>Systematic failure to take independent scientific literature into account | An analysis (Annex 9) of the RAR Volume 1 section 2.0 by the applicants indicates that 95 % of the peer-reviewed scientific literature studies are considered unreliable or supplementary, implying that they have no impact on the risk assessment. Similarly, 93 % of **ecotoxicological studies** were **not taken into account** while scientific literature studies on **endocrine disruption** of glyphosate are considered unreliable. The applicants of the internal review consider that these figures question the application of the Blaise judgment by the RMS and EFSA on the taking | The assessment of the endocrine disrupting properties of glyphosate was conducted following a structured and systematic approach in line with the ECHA/EFSA Guidance (2018) on the hazard identification of endocrine disruptors and thoroughly discussed with the EFSA Working Group on endocrine disruptors. Subsequently, the assessment was extensively discussed at the Pesticides Peer Review Experts' Meeting TC 84.<br><br>All related documents are included in Annex 1 and 2 of the related background documents[45].<br><br>Both regulatory studies and studies retrieved in the peer reviewed open literature throughout the process of the |

[45] Available in the Peer review Report in Open EFSA, Supporting documents section under EFSA-Q-2020-00140, refer to the Peer Review Report: Part 3_Peer Review Report_Glyphosate_Annexes; Peer Review Report_Glyphosate_Annexes_TC84_16 August 2023_EFSA: Annex 2. EFSA ED WG Advice Non-target organisms (NTOs)

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | into account of literature and the opinion of Advocate General Médina (paragraph 108). | renewal of glyphosate were evaluated following the above-mentioned approach. |

**Relevant scientific arguments provided in the review letter submitted by Huglo Lepage for the active substance glyphosate and the conclusions drawn by EFSA on the specific points raised**

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| 1. | AFF. : COLLECTIF DES MAIRES ANTI-PESTICIDES - CRIIGEN C/ COMMISSION EUROPEENNE<br>REF. : 23022229 – CL/BM<br><br>Page 4 | si l'EFSA s'était intéressée aux études universitaires, elle aurait relevé que depuis 2017 la littérature scientifique a foisonné sur le sujet.<br><br>A titre d'illustration, et pour montrer l'envergure mondiale du sujet, en 2017, une étude réalisée en Colombie mettait en lumière l'augmentation du nombre de fausses-couches lors des campagnes d'épandages d'herbicides (Camacho et Mejia, 2017).<br><br>Deux études menées aux Etats-Unis, l'une dans l'Etat de Washington (Caballero et coll., 2018) et l'autre dans le Nebraska (Wan et Lin, 2016), mettent en exergue une augmentation significative du risque de développer la maladie de Parkinson avec l'exposition environnementale au glyphosate.<br><br>Une étude cas-témoins réalisée en Californie (Von Ehrenstein et coll., 2019), a montré une augmentation du risque de troubles du spectre autistiques chez les enfants en lien avec une exposition prénatale à certains pesticides parmi lesquels figure le glyphosate. La même année, une autre étude cas-témoins réalisée en Caroline du Nord a constaté un risque augmenté de malformations cardiaques septales en cas d'exposition prénatale à des pesticides tels que le glyphosate.<br><br>Une étude réalisée en France par un établissement public de recherche médicale, produite en 2021 et que nous détaillerons plus loin, est aussi très documentée sur les risques et dangers du glyphosate. | A robust assessment of all available data has been undertaken in the context of the EU peer review. This included also a comprehensive evaluation of the mammalian toxicology data package. The assessments by the RMS were complemented by the additional expert knowledge provided by the EFSA Working Group, which was subsequently subject to an extensive scrutiny by MSs during the Pesticides Peer Review Experts' Meeting TC 80 before arriving to the final conclusions.<br><br>All public literature submitted to EFSA throughout the regulatory process for the renewal of glyphosate (i.e. included in the RAR or requested during the public commenting phase) was considered as potentially relevant and included in the assessment, including also publications mentioned in the INSERM report. Additional literature identified after the public consultation and considered appropriate to support the assessment was also included. Both studies submitted by the applicants and those retrieved from public literature were equally assessed for their relevance and reliability and were taken into account in a weight of evidence approach.<br><br>The list of the studies assessed including the study appraisal and weight of evidence methodology are described in the Annexes of the Pesticides Peer Review Experts' Meeting Report TC 80.<br><br>More specifically, public literature available to EFSA on neurotoxicity included primary research studies (in vivo, in vitro and mechanistic), reviews, etc, resulting in about 60 neurotoxicity studies |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | Il existe donc un consensus scientifique évident à l'échelle mondiale, qui ne peut raisonnablement pas être nié et dont des conséquences devaient être tirées et refusant de réapprouver le glyphosate.<br><br>La prise en compte réelle de ces études ne permettait pas d'aboutir à la décision prise par la Commission, en raison d'une réalité scientifique sur certains points, et d'un doute suffisamment important sur d'autres, justifiant, au nom du principe de precaution. | (epidemiological studies included). Amongst these, 9 studies were considered for autism, 13 studies for Parkinson's disease and 2 epidemiological studies for amyotrophic lateral sclerosis (ALS). To assess the available literature, the EFSA Working Group followed a structured approach as described in the Annex 7 of the Pesticides Peer Review Experts' Meeting Report TC 80[46], aiming to provide a weight-of-evidence evaluation of the possible effects of glyphosate on human health. Individual studies commented during the public consultation phase were grouped in different sub-sections (e.g., autism, Parkinson's disease, neurotransmitters, developmental neurotoxicity and other neurotoxicity studies).<br><br>From the 7 epidemiological studies available (see Annex 4 of the Pesticides Peer Review Experts' Meeting Report TC 80), including reviews, investigating the possible relationship between exposure to glyphosate and autism, only one found significant associations with glyphosate exposure (von Ehrenstein et al., 2019). However, unambiguous levels of exposure to single pesticides without accounting for other co-exposures was considered hard to assume, and the EFSA Working Group considered that no conclusion could be drawn on the possible correlation between exposure to glyphosate and autism since this unique study has limitations regarding exposure assessment.<br><br>Regarding the epidemiological studies on Parkinson's disease, 8 studies were made available to the EFSA |

[46] available in the Peer Review Report in the Open EFSA, section 'Supporting documents' under EFSA Question number EFSA-Q-2020-00140: https://open.efsa.europa.eu/study-inventory/EFSA-Q-2020-00140); refer to Part 3_Peer Review Report_Glyphosate_Annexes. TC 80.

| No. | Column 1<br>Reference to review letter | Column 2<br>Argument | Column 3<br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | | Working Group but only Caballero et al., 2018 was considered acceptable with restrictions. This study found a significant association with glyphosate exposure, but the limitations inherent to GIS-based exposure assessment prevent from drawing robust conclusions. The remaining studies were case reports, reviews or assessed exposure using an ecological approach (at the group level rather than at individual level).<br><br>Regarding amyotrophic lateral sclerosis (ALS), only two studies were made available with inconsistent results. While the AHS agriculture cohort[47] found no association between glyphosate and ALS, a case-control study (Andrew et al., 2021) reported a significant increased risk based on glyphosate exposure using GIS data (an indirect estimate of actual exposure that pose risk of misclassification).<br><br>Overall, the evidence made available to the EFSA Working Group on neurotoxicity was found limited and quite heterogeneous regarding exposure (most studies used GBHs rather than glyphosate active substance, and at a very wide dose range) as well as in terms of the endpoints assessed.<br><br>In general, the reliability of the studies was considered low to infer causal associations, which limited their utility for risk assessment and no robust conclusions could be drawn.<br><br>Overall, based on the current state of knowledge, the peer review concluded that there was insufficient evidence on the association between glyphosate exposure and autism spectrum disorder (ASD) or |

[47] Kamel F, Umbach DM, Bedlack RS, Richards M, Watson M, Alavanja MC, Blair A, Hoppin JA, Schmidt S, Sandler DP. Pesticide exposure and amyotrophic lateral sclerosis. Neurotoxicology. 2012 Jun;33(3):457-462

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | | amyotrophic lateral sclerosis (ALS) and considered that the integration of human observational studies with the limited experimental evidence from in vitro and in vivo studies does not trigger a concern for parkinsonism.<br><br>In relation to the two papers cited (Camacho and Mejia, 2017 and; Wan and Lin, 2016) it is noted that they can be considered of low relevance for the risk assessment of glyphosate. Camacho and Mejia, 2017, evaluated the effects that the aerial spraying of herbicides to reduce illicit coca cultivation has on health outcomes in Colombia. The paper associated the area where aerial spraying was performed, with short-term health effects reported at individual medical consultations. The type of study does not allow estimating the exposure levels and in general to confirm a causal association between the aerial spraying activities and the onset of human health effects. It is noted however that the conditions of use described in the paper are not comparable with the authorised conditions of use of glyphosate products in the EU. The study was appraised by the RMS and by the EFSA Working Group but it was considered too general to draw any conclusions and was eventually not included in the risk assessment.<br><br>The second paper (Wan and Lin, 2016), establishes a possible association between exposure to a wide series of pesticides including glyphosate and the incidence of Parkinson's disease (PD) at state level (Nebraska, US). The study found significantly increased risk of PD in counties with higher levels of exposure to atrazine, broxomy, alachlor, metribuzin and glyphosate, without however establishing a clear causal association. Co-exposure to multiple |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | | pesticides had stronger correlation to PD incidence than exposure to an individual pesticide. The authors noted a series of potential confounding factors that could have impacted on the analysis and noted that the observed weak regression indicates that pesticide exposure is not the sole contributor to PD, and other environmental and life-style factors may also contribute to the prevalence of PD in Nebraska. The study was appraised by the RMS and by the EFSA Working Group and discussed in the Pesticides Peer Review Experts' Meeting TC 80 due to different conclusions in the study appraisal (the RMS considered it as supportive while it was appraised as not acceptable due to the high risk of bias by EFSA). |
| 2. | AFF. : COLLECTIF DES MAIRES ANTI-PESTICIDES - CRIIGEN C/ COMMISSION EUROPEENNE<br>REF. : 23022229 – CL/BM<br>Pages 5-7<br><br>INSERM | …un rapport de **l'Institut national de la santé et de la recherche médicale** (Inserm), **établissement public à caractère scientifique et technologique français spécialisé dans la recherche médicale**, de près de 1 000 pages consacrés à l'impact des pesticides sur la santé et absolument accablant en ce qui concerne le glyphosate.<br><br>Dans cette étude intitulée « Pesticides et effets sur la santé », l'INSERM affirme : « *Le glyphosate et son métabolite l'AMPA sont des contaminants retrouvés dans les produits alimentaires, des produits agricoles bruts ou des produits transformés* ».<br><br>L'étude consacre un chapitre entier au glyphosate. Ce chapitre évoque un certain nombre de risques sanitaires potentiellement associés à l'exposition professionnelle ou environnementale au glyphosate dont le lymphome non-hodgkinien. Le rapport se réfère à plusieurs méta-analyses l'une par le consortium AHS qui fait apparaître une élévation statistiquement significative du risque, | A rigorous evaluation of the available epidemiological data was performed by the EFSA Working Group on glyphosate and discussed in the Pesticides Peer Review Experts' Meeting TC 80. All public literature submitted to EFSA throughout the regulatory process for the renewal of glyphosate (i.e. included in the RAR or requested during the public commenting phase) was considered as potentially relevant and included in the evaluation. This set of publications included the publications reported in the INSERM report. Additional literature identified after the public consultation and considered appropriate to support the assessment was also included in the assessment. Public literature available to EFSA included primary research studies (case-control studies, cohort studies, etc.), narrative reviews, systematic reviews, meta-analysis, etc. The documents available during the commenting phase, the RAR and its revised version were used as reference documents. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | deux autres méta-analyses de 2016 et 2019 (page 812 du rapport). Le rapport conclut ainsi : « ***en résumé des nouvelles données renforcent la présomption d'un lien entre glyphosate et le risque de LNH dans les populations d'agriculteurs (présomption moyenne). Cette conclusion repose d'une part sur la métaanalyse récemment publiée par le consortium de cohorte d'agriculteurs Agricoh (…) et sur les trois méta-analyses analyses récentes réalisées à partir d'études anciennes montrant systématiquement un risque augmenté. »***<br><br>En dehors du lymphome non-hodgkinien sont évoqués également, de manière non exhaustive:<br><br>- le **lymphome de Hodgkin** : Risque élevé mais à la limite de la signification statistique.<br>Les données n'existaient pas dans les conditions de l'expertise collective de 2013. Aujourd'hui, il existe une présomption de lien entre l'exposition au glyphosate et la leucémie sur la base des résultats de la cohort AHS.<br><br>- Il en va de même des **cancers de la vessie.**<br><br>- **S'agissant des pathologies respiratoires,** le nombre d'études est encore limité mais la plupart des études montre un excès de risque de sifflement allergique ou non et d'asthme.<br><br>- S'agissant de la **maladie de Parkinson,** deux études existent ; l'une conclut à un excès de risque, l'autre à une absence de risque après ajustement à d'autres pesticides. | |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | - Une association également entre **troubles anxiodépressifs** et exposition aux pesticides a été mise en oeuvre pour de nombreuses familles de pesticides sans spécificité particulière pour le glyphosate.<br><br>- S'agissant des **pathologies de la thyroïde**, l'augmentation du risque d'hypothyroïdie a été retenue chez les hommes applicateurs de pesticides.<br><br>- Quelques études témoins mettent également en évidence des anomalies de la grossesse et des **maladies chez les enfants** nés de parents applicateurs de glyphosate.<br>Il s'agit d'études américaines qui mettent en évidence un risque augmenté de survenue de **troubles du spectre autistique** chez les enfants et avec une exposition prénatale à certains pesticides.<br><br>- Il faut également souligner l'existence d'une **maladie rénale** survenue en particulier dans les zones tropicales et notamment au Sri Lanka qui a montré que cette maladie constituait effectivement un important problème de santé publique et qu'elle était lié au facteur d'activité agricole, sans que le rôle spécifique du glyphosate, pourtant largement utilisé, puisse être affirmé.<br><br>- S'agissant des études de c**ancérogenèse**, le rapport de l'Inserm conclut : « les données de cancérogenèse expérimentale entre l'exposition au glyphosate et l'occurrence de quelques types de tumeurs **établissent un lien** généralement limité à un seul sexe et selon les cas, à des lignées sensibles à de très fortes doses d'exposition. Au vu de cet ensemble de résultats, le | |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | niveau de preuve de cancérogénicité chez le rongeur est non nul et relativement limité. De plus, associé un agent initiateur, dispose à présent un effet promoteur à des doses beaucoup plus faibles que celles préalablement testées. Ceci étant, un effet génotoxique en rapport avec un stress oxydatif pourrait avoir un effet de promoteur tumoral ».<br><br>- S'agissant de la **génotoxicité, «** *les différents modèles expérimentaux montrent des* ***résultats positifs in vitro et in vivo*** *(…). En comparaison des niveaux d'exposition, plusieurs tests in vitro observent des effets génotoxiques à des concentrations proches de celles qui peuvent être détectées dans l'environnement. À titre d'exemple, en France, les concentrations de glyphosate ne dépassent pas 0,07 mg dans les eaux de surface ; cette valeur est donc proche de celle induisant des effets génotoxiques sur A sur Oreochromis niloticus. »* Le rapport souligne que les nombreux travaux publiés expriment des résultats plutôt positifs quant à un effet génotoxique ; en revanche les deux essais sont négatifs en ce qui concerne les effets sur la mutagenèse.<br><br>- Le rapport met également en lumière des effets en termes de **cytotoxicité et de toxicité mitochondriale**.<br><br>De plus, le rapport met également en lumière des effets pro oestrogéniques via des récepteurs aux oestrogènes constatés à de fortes mais aussi à de faibles doses. Le rapport considère que le glyphosate pourrait être un perturbateur endocrinien agissant au niveau des fonctions de développement ou/et de reproduction. | |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | Le glyphosate a également un effet épigénétique pour des valeurs inférieures à la NOAEL sur une dose d'exposition courte. Il faut ajouter des effets neurotoxiques. Le rapport indique que « de nombreuses études mettent en évidence des dommages génotoxiques ; s'ils ne sont pas réparés sans erreur par les cellules, peuvent conduire à l'apparition de mutations et déclencher ainsi un processus de cancérogenèse ». | |
| 3. | AFF. : COLLECTIF DES MAIRES ANTI-PESTICIDES - CRIIGEN C/ COMMISSION EUROPEENNE REF. : 23022229 – CL/BM<br><br>Pages 7-8<br>Les failles de la toxicologie réglementaire | Les impacts des pesticides sur la santé, des humains, comme de tous les écosystèmes sont uniquement basés sur la toxicité de la seule « matière active » déclarée par le fabricant… Dans le cas qui nous occupe le glyphosate. Ainsi, l'EFSA, l'Anses, et la Commission européenne considèrent qu'il n'existe aucune différence de toxicité en la molécule déclarée active et le produit utilisé par les agriculteurs et les particuliers.<br>**De ce fait il nous est loisible de répertorier et d'utiliser toutes les études, quelles concernent la molécule déclarée active : le glyphosate (G) ou le produit commercial, c'est-à-dire les Herbicides à Base de Glyphosate (HBG).**<br>Cela nous permet de noter que de nombreux « Roundup » ou « HBG » ont été retirés du marché du fait de leur toxicité importantes due aux coformulants. En effet, en 2016, l'Anses procède au retrait de 132 Roundup associant la substance active glyphosate au coformulants POE-Tallowamine ; en 2019, l'Anses retire du marché 36 produits à base de glyphosate… Tout cela après des années de mise sur le marché et de vente !<br><br>Cela met en cause directement la crédibilité des agences d'autorisation des pesticides… | PPP representative formulations are evaluated as part of the assessment of the active substances. EFSA concurs that studies performed on products other than the representative formulation should not be disregarded a priori as non-relevant since they could potentially provide information as regards the toxicity of the active substance itself or information on potential higher toxicity of that formulation compared to the representative formulation. For this reason, in the case of glyphosate, the applicants were requested to disclose information on the composition of commercial PPPs to allow the assessment of the equivalence with the composition declared for the representative formulation and the interpretation of public literature toxicological and ecotoxicological studies conducted on GBHs. Where available, this information has been provided to the MSs' and EFSA's risk assessors. Additionally, it is worthwhile noting that the available literature on GBHs that were available to the peer review were considered in the weight of evidence of all the assessed endpoints and not only for the assessment of the carcinogenic properties of glyphosate. In addition, it should be stressed that commercial PPPs |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | Nous ne pouvons donc que constater que ces protocoles manquent de pertinence, d'indépendance et de transparence. En effet, ces tests incomplets ainsi que leurs résultats sont réalisés par le fabricant et de surcroit, la composition des coformulants est le plus souvent inconnue, car relevant du secret industriel voire de la propriété intellectuelle du fabricant. A cela s'ajoute le rôle péjoratif des lobbies de l'agrochimie et des tenants de l'agriculture industrielle qui n'est plus à démontrer … (Robin Mesnage et al.2019)[48] : « *Aperçu de la confusion autour des coformulants tensioactifs dans les herbicides à base de glyphosate »* : « La composition des HBG étant légalement classée comme information commerciale confidentielle, la confusion concernant l'identité et les concentrations des coformulants est courante et les descriptions des substances testées dans les études publiées sont souvent erronées ou incomplètes. Afin de dissiper cette confusion, des lois exigeant la divulgation de la composition chimique des produits pesticides pourraient être promulguées ».<br><br>2- **Il est à noter d'autre part, que les agences d'accréditations axent principalement leur regard sur les effets cancérigènes** telle en témoigne la controverse autour du classement du glyphosate « cancérigène probable ». On ne meure pas uniquement du cancer ! Une petite revue des impacts santé du glyphosate (G) et des herbicides à base de glyphosate (HBG) est nécessaire: | containing approved active substances are subject to further evaluation at MS level.<br><br>The cited publication of Mesnage et al. (2019) is a review paper describing narratively the chemical identification and toxicity profile of some co-formulants (notably including co-formulants currently not allowed for use in PPPs in the EU (Polyethoxylated tallowamine surfactants), and their replacements. No original data are presented. The assessment of the toxicological profile of the representative formulation was included in the RAR.<br><br>In relation to the evidence that glyphosate can cause oxidative stress, EFSA concluded that glyphosate may induce oxidative stress as shown in some in vitro and in vivo studies, however increased oxidative stress was not consistently demonstrated in the available studies. It should be also noted that oxidative stress is not a toxicological endpoint *per se*, and it is considered a molecular mode of action by which chemical substances may exert their toxicological properties. Therefore, any effects possibly mediated by oxidative stress (e.g. DNA damage, or increased tumour incidence, *inter alia*) would have been identified considering the extensive toxicological dataset available for glyphosate. |

[48] Robin Mesnage et al .2019. **Insight into the confusion over surfactant co-formulants in glyphosate-based herbicides.** Review Food Chem Toxicol. 2019. doi: 10.1016/j.fct.2019.03.053

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | **Impacts sur les systèmes** : - **endocrine** (Juan P Muñoz et al. 2021)[49] ; - **le stress oxydatif** favorisant le développement de l'inflammation chronique et donc potentiellement des cancers (Vicky C Chang et al. 2023)[50]. (Xiaojing Wang et al. 2022)[51] ; - **immunitaire** (Ambra Maddalon et al. 2022)[52], (Ewing Duque-Díaz et al. 2022)[53]; - cardiaque (Jian Lu et al 2023)[54] - **nerveux** (Joanna K Winstone et al. 2022.)[55]; - **le microbiote colique** (Peter C Lehman et al. 2023)[56]; et bien d'autres encore … | |
| 4. | AFF. : COLLECTIF DES MAIRES ANTI-PESTICIDES - CRIIGEN C/ COMMISSION EUROPEENNE REF. : 23022229 – CL/BM<br><br>Environment and Biodiversity – page 8 | 3- **Les impacts écosystémiques environnementaux sont quasiment passés sous silence:**<br>impacts sur la faune, la flore, pollution de l'eau, de l'air, des sols, des aliments, développement de résistance au glyphosate nécessitant de cumuler deux herbicides (ex : 2-4, D et glyphosate)<br>... Ainsi que les coûts externalisés que cela engendre… | EFSA disagrees that impacts on the environment were ignored. According to the provisions of Regulation 1107/2009, data requirements of the Regulations 283/2013 and 284/2013 and the Uniform Principles (Regulation 546/2011), a risk assessment was carried out for non-target organisms i.e. birds and wild mammals, aquatic organisms, non-target arthropods and bees, soil organisms and non-target terrestrial plants from the representative uses supported in the renewal process. These risk assessments are aimed at predicting the likelihood of effects from these uses, based on the hazard of the substance and predicted |

[49] Juan P Muñoz et al. 2021. **Glyphosate and the key characteristics of an endocrine disruptor: A review. Review Chemosphere.** 2021. doi: 10.1016/j.chemosphere.2020.128619
[50] Vicky C Chang et al. 2023. Glyphosate exposure and urinary oxidative stress biomarkers in the Agricultural Health Study. J Natl Cancer Inst. 2023. doi: 10.1093/jnci/djac242
[51] Xiaojing Wang et al. 2022**. Oxidative Stress and Metabolism: A Mechanistic Insight for Glyphosate Toxicology.** Review Annu Rev Pharmacol Toxicol. doi: 10.1146/annurev-pharmtox-020821-111552
[52] Ambra Maddalon et al. 2022. **Direct Effects of Glyphosate on In Vitro T Helper Cell Differentiation and Cytokine Production.** Front Immunol. doi: 10.3389/fimmu.2022.854837
[53] Ewing Duque-Díaz et al. 2022**. Glyphosate, AMPA and glyphosate-based herbicide exposure leads to GFAP, PCNA and caspase-3 increased immunoreactive area on male offspring rat hypothalamus.** Eur J Histochem. 2022. doi:10.4081/ejh.2022.3428
[54] Jian Lu et al 2023**. Glyphosate Causes Vascular Toxicity through Cellular Senescence and Lipid Accumulation.** Chem Res Toxicol. 2023. doi: 10.1021/acs.chemrestox.3c00116.
[55] Joanna K Winstone et al. 2022. **Glyphosate infiltrates the brain and increases pro-inflammatory cytokine TNFα: implications for neurodegenerative disorders.** J Neuroinflammation. doi: 10.1186/s12974-022-02544-5.
[56] Peter C Lehman et al. 2023. **Low-dose glyphosate exposure alters gut microbiota composition and modulates gut homeostasis.** Environ Toxicol Pharmacol. 2023. doi: 10.1016/j.etap.2023.104149.

| No. | Column 1<br>Reference to review letter | Column 2<br>Argument | Column 3<br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | | exposure from each representative use. It is noted that as part of the environmental risk assessment, fate and behaviour in different matrices (i.e. soil, water, aquatic sediment, treated drinking water and air) was also investigated along with the prediction of the concentrations of residues in soil, water and aquatic sediment for each representative use. As part of the human consumer risk assessment, residues in food commodities from each representative use was quantified and assessed.<br>Environmental risks from exposure to combination of several substances (other than technical mixtures of active substance(s) and their co-formulants undergoing an authorisation procedure) are outside of the scope of Regulation (EC) No 1107/2009 and thus of the prospective environmental risk assessment. A consideration of the potential for weeds to develop resistance as a consequence of the representative uses and management measures to avoid this was made.<br><br>It is also noted that the applicants of glyphosate submitted an assessment of biodiversity and effects due to trophic interaction following the provision reported in Annex 1 of Commission Implementing Regulation (EU) No 2017/2324 for MSs to pay particular attention to *the risk to diversity and abundance of non-target terrestrial arthropods and vertebrates via trophic interactions.* A summary of this assessment is reported in the RAR, Volume 3 – B.9 (PPP), B.9.14.1 *Assessment of risk to biodiversity via indirect effects and trophic interactions*[1]. |

| No. | Column 1<br>Reference to review letter | Column 2<br>Argument | Column 3<br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | | The assessment was extensively discussed at the Pesticides Peer Review Experts' Meeting TC 82 (expert consultation point 5.25[2]).<br><br>[1] available in the Open EFSA, 'Supporting documents' section under EFSA Question number EFSA-Q-2020-00140: https://open.efsa.europa.eu/study-inventory/EFSA-Q-2020-00140 (refer to Glyphosate_Final RAR_public.zip)<br><br>[2] available in the Peer Review Report in the Open EFSA, 'Supporting documents' section under EFSA Question number EFSA-Q-2020-00140: https://open.efsa.europa.eu/study-inventory/EFSA-Q-2020-00140 (refer to Part 3_Peer Review Report_Glyphosate_expert meeting reports_public.pdf (TC 82, Expert consultation point 5.25) |

## Relevant scientific arguments provided in the review letter submitted by Aurelia Stiftung for the active substance glyphosate and the conclusions drawn by EFSA on the specific points raised

*It should be noted that the original request for internal review was provided in German language. Where available, a complimentary English translation has been provided to EFSA and ECHA by the Commission for the purpose to facilitate assessment by the Agencies. The English translation as displayed in column 2 has been generated by using an automated machine translation tool. Therefore the quality and accuracy of the translation may vary from the original text and should not be regarded as official translation. Only the original text of the request submitted in German should be considered as the authentic text.*

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| 1. | Approval of the active substance glyphosate – Commission Implementing Regulation (EU) 2023/2660 of 29 November 2023 Request for review under Article 10 of Regulation (EC) No 1367/2006<br><br>Document Number: 867989<br><br>Pages 23 – 30 (EN)<br>[Pages 26-34 (DE)] | **II Insufficient risk assessment due to missing or inadequate guidelines**<br><br>**2. Missing, incomplete or inadequate guidelines for risk assessment**<br><br>**a) Biodiversity**<br><br>The Commission and EFSA note that the necessary methods and guidance on indirect impacts on biodiversity are missing at EU level for risk assessment. Such methods and guidelines still need to be developed in order to allow establishing indirect effects of plant protection products containing glyphosate.<br><br>However, the examination of such effects, which was necessarily carried out in the absence of such a guideline, could not be carried out satisfactorily, inter alia because the data were insufficient.<br><br>**IV Individual analysis of the assessment failures and errors**<br><br>**1.Insufficient assessment of indirect impacts on biodiversity**<br><br>(….)<br><br>The need to take indirect effects on biodiversity into account cannot be contested by the fact that no uniform | The lack of a harmonised approach, as mentioned in EFSA's Conclusion (EFSA, 2023), does not prevent a scientific assessment of indirect effects. Indeed, an assessment was provided by the applicants and extensively discussed during the peer review.<br><br>However, an approach, based on agreed specific protection goals, will give indication on what is the level of protection, and clarify the data requirements and the evaluation / decision making criteria. The current lack of such robust approach, particularly of agreed specific protection goals, prevents risk assessors from identifying and quantifying risks and from drawing any firm conclusion on their acceptability.<br><br>Indeed, the concept of "unacceptable effects" on the environment, as prescribed in Art. 4(3)(e)(iii) of Regulation (EC) No 1107/2009, is in itself rather generic and implies quality judgements which pertain to the sphere of responsibility of the risk managers. In the absence of the setting of specific protection goals clarifying the concept of what is deemed unacceptable, EFSA is in turn not in a position to identify any specific scientific methods to assess indirect effects.<br><br>Furthermore, no provision under the Uniform Principles explicitly empowers EFSA to adopt binding guidance for the definition of criteria or standards to be applied in the context of its scientific |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | Biodiversity / Insufficient assessment of indirect impacts on biodiversity | EU-wide methods and guidelines for assessing such effects have yet been adopted.<br><br>There is no legal obstacle to taking into account indirect effects on biodiversity without the existence of harmonised methods across the EU. The wording in Article 4(3)(e) of Regulation (EC) No 1107/2009 '*where the scientific methods accepted by the Authority to assess such effects are available'* cannot be understood as meaning that the existence of such methods recognised by EFSA is a prerequisite for taking such effects into account, based on the history, … and scheme of the standard.<br><br>An assessment of indirect effects as part of the decision to renew the approval of the active substance would also be technically possible. For example, the interim method already presented by Germany could have been used for the assessment of biodiversity on a transitional basis, as suggested, inter alia, by the German Federal Ministry of Food and Agriculture.<br><br>Even if the European Commission did not consider these and comparable methods to be sufficiently suitable for comprehensible reasons, it should have taken into account the indirect effects on biodiversity when deciding on the renewal of approval, in such a way that the active substance cannot be re-approved. | evaluations, or in the context of the scientific evaluations of the Rapporteur Member States.<br><br>Therefore, it is clear that EFSA may not proactively implement guidance documents and must react to a mandate received from the Commission to develop any guidance and to the endorsement at the SCoPAFF to apply it.<br><br>Overall, considering the complexity of the topic, a robust approach is deemed necessary to ensure that indirect effects are properly introduced and implemented. To this purpose EFSA considers, as priority, the definition and agreement of specific protection goals for non-target arthropods and non-target terrestrial plants. Indeed, arthropods and wild plants are fundamental entities of food networks, and preserving both their biomass and diversity is pivotal to safeguard ecosystem services delivery and ecological function such as habitat provision, food web support, pest control, pollination.<br><br>The implementation of any 'interim solution' without a large consensus and agreement from European Commission and MSs, is therefore not an option that could have been considered. |
| 2. | Honeybees, solitary bees and bumble bees | **II Insufficient risk assessment due to missing or inadequate guidelines**<br><br>**2. Missing, incomplete or inadequate guidelines for risk assessment** | It is noted that the recently published EFSA Bee guidance (EFSA, 2023)[63] is not a draft. Nevertheless, the commenter rightly summarised some of the aspects of this document. Indeed, this guidance was not used for the risk assessment for bees of glyphosate; neither for direct, nor for indirect effects. |

[63] https://www.efsa.europa.eu/en/efsajournal/pub/7989

| No. | Column 1<br>Reference to review letter | Column 2<br>Argument | Column 3<br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | Pages 24-25 (EN)<br>[Pages 27-28 (DE)] | **b) Honeybees, solitary bees and bumble bees**<br>Nor can the effects on bees be conclusively assessed in the PPP authorisation procedures. The required 'Bee guidance' based on the current state of knowledge has still not been adopted. It has been under discussion for years in the relevant committee at EU level (SCoPAFF). There is only an updated draft from EFSA[57], based on the mandate given to EFSA in 2013 (!), following serious gaps identified in the previously used assessment scheme and the need to drastically restrict the approval of several neonicotinoids due to the acute risks to bees[58].<br>Given the timing, this current version of the (draft) guideline cannot yet be taken into account in the risk assessment for glyphosate; for this reason alone, the risk assessment for bees is not up to date with current scientific knowledge.<br>In addition, the (draft) guideline excludes significant risks that would have to be assessed in particular in the case of glyphosate as herbicide:<br>It is true that the (draft) guidance document correctly states that the use of plant protection products can also have an indirect impact on bees, in particular when using herbicides, which reduce the habitat of bees and/or food availability. Although such ecological impacts are relevant, the guidance only considers | Nevertheless, still a robust risk assessment was performed for bees, considering both the data submitted by the applicants and the data retrieved from the open literature using the most comprehensive methodology that was available at the date of the submission of the supplementary dossiers. Key aspects of the risk assessment were discussed and agreed at the Pesticides Peer Review Experts' Meeting TC 82. Therefore, EFSA disagrees with the statement of the commenter that 'The risk assessment carried out for glyphosate therefore has no solid basis'. |

[57] EFSA, Revised guidance on the risk assessment of plant protection products on bees (Apis mellifera, Bombus spp. And solitary bees, 30.03.2023 (approved), veröffentlicht am 11.05.2023; https://efsa.onlinelibrary.wiley.com/doi/10.2903/j.efsa.2023.7989

[58] In particular, the shortcomings of the risk assessments and the underlying assessment scheme on the basis of which these bee-dangerous active substances were approved are subject to several procedures before the Union. Cases C-499/18 and T-429/13

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | direct, toxicity-related effects. With regard to the indirect and ecological impact, reference is made to future EFSA guidance documents on the risk assessment of plant protection products (on the ecosystem service "food web support" for bees).[59]<br><br>In addition, EFSA explicitly confirms that bees can typically be exposed to multiple residues (e.g. mixtures of insecticides, fungicides and herbicides) on agricultural fields, both spatially and temporally. It has been known for many years that representative honey analyses often show a whole cocktail of different active ingredients and plant protection products to which bees are exposed.<br><br>However, the (draft) guideline explains that it does not address the risk assessment of combinations of more than one active substance or plant protection product; risks are also excluded when plant protection products are applied consecutively within a season[60].<br><br>There is also considerable uncertainty and need for further research regarding the risk for bees when plant protection products are applied to flowering plants visited by bees for foraging, which EFSA has identified as relevant. Recommendations to address the knowledge gaps are currently being developed. [61]<br><br>The (draft) guideline also points out that there are currently no internationally recognised test protocols for | |

59 EFSA, op. cit., Chapter 4 ('problem formulation', 1. First paragraph).
60 EFSA, op. cit., Chapter 2
61 EFSA, op. cit., Chapter 4.3.2. and Annex D.

| No. | Column 1<br>Reference to review letter | Column 2<br>Argument | Column 3<br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | the risk assessment with regard to acute and chronic toxicity effects on solitary bees and bumble bees.[62]<br>There is no doubt that a proper risk assessment for bees requires guidance covering all the points that need to be assessed.<br>However, there is no such one to date. The risk assessment carried out for glyphosate therefore has no solid basis. In addition, Member States still lack the basis to compensate for this deficit in the evaluation of plant protection products at national level. For further reference on insufficient consideration of risks for bees, solitary bees and bumble bees, see C.IV below. 2.b). | |
| 3. | Insufficient assessment and consideration of effects on bees and other insects<br>Pages 30-32 (EN)<br>[Pages 34-36 (DE)] | **IV Individual analysis of the assessment failures and errors**<br>**2.Insufficient assessment and consideration of effects on bees and other insects**<br>In addition, new findings that glyphosate has highly toxic direct effect on arthropods, including insects and spiders, even at concentrations well below the permitted amount of spraying, have not been sufficiently taken into account.<br>A study published in early 2023 by the Swiss Federal Institute of Technology University (ETH) Zurich and the German Federal Agency for Nature Conservation (BfN) in the renowned international journal 'Science of the Total Environment' shows that a glyphosate-based | Among the publications mentioned in the document, '*Defarge et al., Science of the Total Environment 865 (2023) 161158'* was screened for potential impact on the risk assessment during the peer review. Based on the criteria for relevance agreed at the Pesticides Peer Review Experts' Meeting TC 82 (see experts' consultation point 5.10 [70]), the study was categorised as 'potentially relevant but insufficient information', since not enough details were available to ascertain the formulation composition used (WeatherMax). Therefore, the study was not used for the risk assessment of glyphosate.<br>Regarding the potential adverse effects after oral ingestion, EFSA has acknowledged that oral exposure is a relevant exposure route for non-target arthropods and that the current risk assessment for (European Commission, 2002) does not |

62 EFSA, op. cit., Chapter 6.1.1.

[70] Refer to the report of the Pesticides Peer Review Experts' Meeting TC 82, available in the Peer Review Report in the Open EFSA, Supporting information section under EFSA-Q-2020-00140 , refer to Part 3_Peer Review Report_Glyphosate_expert meeting reports_public.pdf (TC 82)

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | herbicide seriously harms lacewing larvae directly through dietary intake.<br><br>— Defarge et al., Science of the Total Environment 865 (2023) 161158, (Annex A.15).<br><br>However, when testing possible effects of glyphosate-based herbicides on insects, the dietary intake of substances is not currently examined in the context of approval of active substances, but the insects are only placed on treated areas. However, in the field, insects are very likely to also ingest glyphosate-containing products via diet. The risk posed by the direct effect after oral ingestion is of great importance for environmental safety and reveals a gap in the existing risk assessment.<br><br>There are also indications that glyphosate-containing products negatively influence the behaviour, growth, development, metabolic processes and immune defences of various bee species. A 2021 meta-analysis study found that most bee species, including wild bees and solitary bees, suffer significant adverse effects when exposed to glyphosate.<br><br>— Battisti et al, Is glyphosate toxic to bees? A meta-analytical review, 2021 [64] , PAN, Glyphosate & Glyphosate based herbicides & their impact on bee's health (Annex A.16) | include oral uptake explicitly (EFSA PPR Panel, 2015). [71] However, it was also acknowledged that test guidelines for testing oral exposure to PPP residues on food items are currently lacking.<br><br>The publication quoted in the document '*Battisti et al, 2021. Is glyphosate toxic to bees? 202115, PAN*,' was considered for the peer review, however, since this publication is a review paper it was not deemed to be relevant as it does not provide data from primary source (nevertheless, many of the underlaying data of this paper were anyway captured and considered in the peer review).<br><br>Potential effect on gut microbiome of bees was intensively investigated and assessed (among many other publications, the Motta *at.al.*, 2018 was also considered); the overall outcomes of the assessments are reported in the EFSA Conclusion (EFSA, 2023).<br><br>The second publication referring to bees '*Weidenmüller. et.al. (2022). Glyphosate impairs collective thermoregulation in bumblebees. Science (New York, N.Y.). 376. 1122– 1126'* was not considered since it was outside of the time period for the literature search. However, it was captured by EFSA. After the experts' discussion the RMS has evaluated the study according to the reliability criteria agreed during the Pesticides Peer Review Experts' Meeting TC 82 and the study was not deemed to be reliable.<br><br>The publication by Kiefer et al. (2021) was assessed during the peer review. The publication was considered not relevant for the renewal of glyphosate because the concentration/dose used in |

[64] https://www.sciencedirect.com/science/article/abs/pii/S0048969721004654?via%3Dihub

[71] EFSA PPR Panel (EFSA Panel on Plant Protection Products and their Residues), 2015. Scientific Opinion addressing the state of the science on risk assessment of plant protection products for non-target arthropods. EFSA Journal 2015;13(2):3996, 212 pp. doi:10.2903/j.efsa.2015.3996

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | Since 2018, glyphosate has been known to disrupts the intestinal flora of honey bees. The study 'Glyphosate perturbs the gut microbiota of honey bees' found that honey bees' intestinal microbiome is perturbed after exposure to glyphosate during and after intestinal colonisation. Contact with glyphosate at the early stage of intestinal colonisation increases the mortality of bees when exposed to a pathogen.[65]<br><br>The insect researcher Anja Weidenmüller (University of Konstanz) and her team demonstrated harmful effect of glyphosate on bumble bees in 2022. According to the results of the study 'Glyphosate impairs collective Thermoregulation in bumblebees', the herbicide can cause serious disturbance to the brood development of bumblebees and impact their reproduction. Bumblebees, under the influence of glyphosate and in lack of food, can no longer heat their nest to the required minimum temperature. Sufficient heat is the most important factor in brood development. The brood development is slower as a result of the influence of glyphosate and the colony is at risk of dying.[66] This study is attached as Annex A.17 [10]. | the study was not representative for any of the intended uses of glyphosate. |

[65] Motta, E.V.S, Raymann, K., Moran, N.A. (2018): Glyphosate perturbs the gut microbiota of honey bees. Proceedings oft he National Academy of Sciences (PNAS). 115 (41). 10305-10310, S. 10305. https://www.pnas.org/doi/full/10.1073/pnas.1803880115

[66] Weidenmüller, A., Meltzer A., Neupert, S., Schwarz, A., Kleineidam, C. (2022). Glyphosate impairs collective thermoregulation in bumblebees. Science (New York, N.Y.). 376. 1122-1126. https://www.science.org/doi/10.1126/science.abf7482

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | — Weidenmüller. et.al. (2022). Glyphosate impairs collective thermoregulation in bumblebees. Science (New York, N.Y.). 376. 1122– 1126[67] (Annex A.17)<br><br>As with bees, glyphosate damages the beetle biome. The symbiotic bacteria necessary for the formation of the exoskeleton (cuticle) are damaged by glyphosate.[68] This has been demonstrated in 2021 by researchers from Johannes Gutenberg University Mainz, the Max Planck Institute for Chemical Ecology in Jena and the National Institute of Advanced Industrial Science and Technology in Japan in cereal leaf beetles in the study 'Inhibition of a nutritional endosymbiont by glyphosate abolishes mutualistic benefit on cuticle synthesis in Oryzaephilus surinamensis.[69]<br><br>In order to reduce the risk to bees, bumble bees and other pollinators, it was at least necessary, under Article 20(1)(a) of Regulation (EC) No 1107/2009, to prohibit the use of glyphosate in flowering plants or at least significantly restrict it. | |

[67] The study is considered particularly valuable by the internationally well-known bee researcher and neurobiologist *Randolph Menzel* (Free Uni-Versität Berlin) because it is of high quality method and also allows statements on the real conditions for wild bees, cf. Podbregar, Nadja: Glyphosate colds bumble bees. Herbicide disrupts breeding and active heat production in bumble bees, in: Scinexx, the Knowledge Magazine, 3 June 2022, https://www.scinexx.de/news/biowissen/glyphosat-macht-hummeln-kalt/ https://www.scinexx.de/news/biowissen/glyphosat-macht-hummeln-kalt/

[68] Dr Engel, Tobias: Glyphosate inhibits symbiotic bacteria from beetles. The pesticide damages the microorganisms necessary for the formation of the outer skeleton of cereal leaf beetle, in: Max Plank Society, 11.5.2021, https://www.mpg.de/16860036/0506%E2%80%90choe%E2%80%90die%E2%80%90achillesferse%E2%80%90eines%E2%80%90kaefers%E2%80%90155371%E2%80%90x (accessed on 25 January 2023).

[69]Kiefer, J. S. T., Batsukh, S., Bauer, E., Hirota, B., Weiss, B. Wierz. J. C., Fukatsu, T., Kaltenpoth, M., Engl, T. (2021). Inhibition of a nutritional endosymbiont by glyphosate abolishes mut ualistic benefit on cuticle synthesis in *Oryzaephilus surinamensis*. Communications Biology. 4. https://pubmed.ncbi.nlm.nih.gov/33976379/

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| 4. | Insufficient assessment of the impact on mammals<br><br>Page 32 (EN)<br>[Page 36 (DE)] | **IV Individual analysis of the assessment failures and errors**<br><br>**3.Insufficient assessment of the impact on mammals**<br><br>EFSA identified, inter alia, a long-term risk for small herbivorous mammals for some uses which, in the view of the applicant's [*of the current review*], should have led to the refusal of re-approval.<br><br>The delegation of the assessment of this risk to the Member States, as provided for in Implementing Regulation (EU) 2023/2660, is incorrect for the reasons set out above [*cf also section III on page 29*].<br><br>In addition, EFSA itself notes that risks to non-target wild terrestrial vertebrate animals have been identified as a result of exposure to a glyphosate product (representative formulation) (EFSA, Peer Review, p. 25). | The document (in column 2) highlights that, for some of the uses assessed in the renewal process, EFSA concluded a high long-term risk to mammals. This is consistent with the EFSA Conclusion. |
| 5. | Insufficient assessment of impacts on surface waters<br><br>Contamination of surface waters<br><br>Pages 32-35 (EN) | **IV Individual analysis of the assessment failures and errors**<br><br>**4. Insufficient assessment of impacts on surface waters**<br><br>**a) Contamination of surface waters**<br><br>The renewal decision does not take sufficient account of the high contamination of surface waters by glyphosate and AMPA. Contrary to the EU Commission's assessment, the concentrations of glyphosate and its degradation products to be expected in water bodies must be qualified as unacceptable and preclude approval. | The aquatic risk assessment for glyphosate and AMPA adhered to the EFSA PPR Panel Guidance Document On Tiered Risk Assessment For Plant Protection Products For Aquatic Organisms In Edge-Of-Field Surface Waters In The Context Of Regulation (EC) No 1107/2009 (EFSA Journal 2013;11(7):3290, 268 pp. doi:10.2903/j.efsa.2013.3290), ensuring that the risk assessment process was conducted in accordance with established EU standards and methodologies specific to aquatic environments. There are no current environmental quality standard (EQS) levels for glyphosate and AMPA at the European level in place. Any proposed EQS values at the European level are still under discussion and not implemented. This is the reason why the peer review utilised the regulatory acceptable |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | [Pages 36-40 (DE)] | With regard to the spread of the active substance glyphosate and its degradation products in surface waters, the European Commission relies on the fact that in the context of public surface water monitoring, the regulatory acceptable concentrations (RAC) for glyphosate and AMPA were complied with in a very high proportion of samples (around 99 %) (EFSA, Peer Review, p. 20; Combined dRR Volume 3 – B.8, p. 14).<br><br>However, the RAC values of 400 µg/L and 100 µg/L used here are far too high to assess the acceptability of environmental risks. In particular, they are significantly above current and planned environmental quality standards.<br><br>Directive (EU) 2020/2184 on the quality of water intended for human consumption (Drinking Water Directive) sets in its Annex I a limit value of 0.1 µg/l for pesticides and relevant metabolites. Compliance with this value must be ensured not only for drinking water, but also for surface waters currently or in the future used for the abstraction of drinking water. Under Article 7(3) of Directive 2000/60/EC establishing a framework for Community action in the field of water policy (Water Framework Directive – WFD), Member States are to ensure the necessary protection of the bodies of water identified in accordance with paragraph 1 of that provision which are used for (current/future) drinking water extraction 'in order to prevent deterioration of their quality and thus reduce the amount of treatment required for the abstraction of drinking water'. Such deterioration, which increases the amount of treatment, occurs when the drinking water parameters of the | concentrations (RAC) values for glyphosate and AMPA for comparison to water concentration monitoring data.<br><br>Monitoring results from public surveys cannot be assimilated to concentrations that can be used for regulatory exposure assessment and be assessed against a regulatory exposure assessment goal without additional information (e.g. aspects such as agricultural context, including farmer usage of plant protection products, or site characterisation).<br><br>Furthermore, the comparison of surface monitoring data against the threshold of 0.1 µg/L is based on measured concentrations in raw surface water, before any water treatment process. This kind of assessment is not a regulatory requirement under Regulation (EC) No 1107/2009 but it is provided only to support biodiversity considerations.<br><br>It is also worth noting that the available comparison of the surface water monitoring data against the threshold of 0.1 µg/L is based on measured concentrations in raw surface water, before any water treatment process (the proportion of sampling locations potentially intended to supply drinking water is unknown). This kind of assessment is not a regulatory requirement under Regulation (EC) No 1107/2009. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | drinking water directive have been exceeded.72 The precautionary protection of drinking water resources required by Article 7 of the WFD therefore requires that the specific drinking water limit value of 0.1 µg/L for glyphosate at least in the water used for the production of drinking water of today/future is complied with.<br><br>According to the available monitoring data, this is not the case for a significant proportion of the monitoring sites. The evaluation of the monitoring data from the GRG concluded that in approximately 23 % of the samples (54.0 % of the sites) values above the threshold value of 0.1 µg/L were measured. For the metabolite AMPA, even 47.5 % of the measurements found exceedances of the threshold values (Combined dRR, Volume 3 – B.8, p. 14, 252).<br><br>In Germany, glyphosate and AMPA are detected nationwide at around 40 % to 60 % of the surveyed sampling points in concentrations above 0.1 µg/L.<br><br>— *Bund-Länder-Arbeitsgemeinschaft Wasser (LAWA), Micropollutants in Waters, 2016, p. 7 (Annex A.18).*<br><br>It should be borne in mind that relatively few monitoring data are available. It is likely that the monitoring data collected by the GRG represent only a fraction of the actual contamination. The Renewal Regulation should at least have laid down binding provisions to improve the dataset.<br><br>Regulation (EC) No 1107/2009 stresses at several points the need to ensure consistency with the requirements of the WFD (recitals 16, 47, Article 44). | |

72 See Opinion of the Advocate General of 2 March 2023, C-723/21, Celex No 62021CC0723, point 102.

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| 6. | Insufficient assessment of impacts on surface waters<br><br>Effects on aquatic organisms<br><br>Impact on non-target aquatic plants<br><br>Pages 35-36 (EN)<br><br>[Pages 40-41 (DE)] | **IV Individual analysis of the assessment failures and errors**<br><br>**4. Insufficient assessment of impacts on surface waters**<br><br>**b) Effects on aquatic organisms**<br><br>The high level of water pollution is also unacceptable because unacceptable ecotoxicological effects from glyphosate on aquatic organisms cannot be ruled out.<br><br>Glyphosate has been identified in the CLP Regulation as toxic to aquatic life with long-lasting effects (Aquatic Chronic 2; H411). There are studies showing that glyphosate can be toxic to aquatic organisms at lower concentration levels.<br><br>*— Uren Webster & Santos, 2015 (**Annex A.19**)*<br><br>Amphibians are particularly sensitive to glyphosate and glyphosate containing products.<br><br>Several studies also indicate that the use of glyphosate is partly responsible for the sharp decline in amphibians.<br><br>*— Plötner & Matschke 2012 (**Annex A.20**), Wagner & Lötters 2013 (**Annex A.21**)*<br><br>**c) Impact on non-target aquatic plants**<br><br>There is also scientific evidence that glyphosate can be toxic for macrophyte communities (large aquatic plants). | 4b) The CLP classification of glyphosate (ECHA, RAC Opinion, 2022) mentioned by the document is based on the same fish endpoint (NOEC = 1 mg/L) which is driving the aquatic risk assessment in the EFSA Conclusion.<br><br>Among the papers mentioned in the document (column 2), one (Uren Webster and Santos, 2015) was already considered and evaluated during the peer review process. Another one (Wagner and Lötters, 2013) was excluded at the screening level, as it is an expert opinion containing no additional data. Finally, Plöttner and Matschke (2013) was not captured in the systematic literature search provided by the applicants, but as this is a review, it does not provide additional data that may be expected to alter the outcome of the risk assessment.<br><br>4c) The only study mentioned by the document is Sesin et al. (2021). This study was extensively considered in the peer review[73] and triggered the conclusion for a data gap to address the risk to aquatic macrophytes due to contact exposure via spray drift. This further resulted in an assessment not finalised. |

[73] See expert consultation point 5.14 in the report of the Pesticides Peer Review Experts' Meeting TC 82, available in the Peer Review Report in the Open EFSA, Supporting information section under EFSA-Q-2020-00140 , refer to Part 3_Peer Review Report_Glyphosate_expert meeting reports_public.pdf (TC 82)

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | *— Sesin et al, 2021 (Annex A.22)*<br><br>EFSA concluded that the risk assessment for aquatic macrophytes due to exposure to spray drift could not be finalised since Further information to investigate the risk for aquatic macrophytes due to contact exposure via spray drift is needed, including an assessment of the toxicity of the active substance and the formulation to standard macrophytes species via this route of exposure (EFSA, Peer Review, p. 31).<br><br>As explained above, this data gap already requires the non-renewal of the approval of the active substance. | |
| 7. | Insufficient assessment of the impact on groundwater<br><br>Pages 36-37 (EN)<br>[Pages 41-42 (DE)] | **IV Individual analysis of the assessment failures and errors**<br><br>**5. Insufficient assessment of the impact on groundwater**<br><br>Unacceptable effects on groundwater cannot be ruled out either.<br><br>Such unacceptable effects shall be assumed if Glyphosate and/or its metabolites consequent to the use of glyphosate-containing products leaches to groundwater at concentrations > 0.1 µg/L.<br><br>EFSA has identified a data gap with regard to groundwater exposure via bank infiltration and connectivity of surface water bodies to groundwater aquifers, that was considered relevant for all representative uses (EFSA, Peer Review, pp. 20, 39). This data gap, which points to a significant risk to groundwater, is an obstacle to the approval of the active substance. | The peer review acknowledged that the widespread use of glyphosate makes it essential to consider potential routes of groundwater exposure, particularly through bank infiltration and the connectivity between surface water bodies and groundwater aquifers. The data gap identified in the EFSA Conclusion highlights the need for further research and data collection to fully understand and address this potential route of exposure. However, as a matter of fact, there are locations in Europe where the movement of water from surface water bodies to groundwater (or vice versa) is limited or minimal due to the specific characteristics of the landscape and hydrological processes. In these areas, the risk of contamination of groundwater from substances like glyphosate, which might be present in surface water bodies, may not be a significant concern. Therefore, this data gap is not "an obstacle to the approval of the active substance" because safe uses could be identified by risk managers.<br><br>The risk of groundwater exposure from glyphosate application on sealed and very permeable surfaces was not identified by the EFSA peer review, as such uses were not included in the representative uses applied for and peer reviewed in the context |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | EFSA has also identified an increased risk of groundwater exposure from glyphosate application on sealed and very permeable surfaces (EFSA, Peer Review, p. 20).<br><br>With regard to the modelling results for the remaining exposure routes, which, according to EFSA, show a low risk of exceeding the 0.1 µg/L threshold (EFSA, Peer Review, p. 19), it should be noted that the groundwater scenarios used for modelling are partly not representative. For example, sensitive areas, such as karstic catchment areas, with sometimes very limited overlaying powers, are not depicted in the groundwater scenarios and are therefore not taken into account in the risk assessment.<br><br>In addition, monitoring data indicating an exceedance of 0.1 µg/L threshold value were incorrectly assessed and not fully taken into account.<br><br>Even in the monitoring data collection submitted by the GRG, exceedances of the threshold value of 0.1 µg/L were found (0.6 % for glyphosate and 0.7 % for AMPA, Combined dRR Volume 3 – B.8, p.).<br><br>The relatively low proportion of exceedances does not alter their relevance.<br><br>Moreover, this collection of public monitoring data provided by the GRG is incomplete. For example, monitoring data for Germany for the period 1996-2008 were used in the GRG collection for Germany (Combined dRR Volume 3 – B.8, p. 8), although more recent nationwide data from 2019 are available in Germany for the period 2013-2016. According to this, glyphosate occurs in the 20th place in the ranking of the most frequently detected PPP active substances or | of the renewal application. The indication of an increased risk in the column 2 seems to be an incorrect citation of the EFSA conclusion. EFSA understands that this was rather identified by the European Commission in their review report, that was developed by them and risk managers after the finalisation of the EFSA conclusion.<br><br>With regard to the modelling, the groundwater exposure assessment was performed according to the current guidance document (European Commission, 2014. Assessing potential for movement of active substances and their metabolites to ground water in the EU. Report of the FOCUS Workgroup. EC Document Reference SANCO/13144/2010-v. 3, 613 pp, as outlined in Generic guidance for tier 1 FOCUS groundwater assessment, v. 2.2 May 2014). Predicted Environmental Concentrations have been calculated for the realistic worst-case groundwater FOCUS scenarios, representing the majority of the agricultural and climatic conditions found across the EU. It is acknowledged that in areas where soil layers sit above karstic geology and are shallower than 1 meter, the FOCUS groundwater scenarios may not adequately address leaching. Therefore, assessment for national product authorisations would be necessary, and there may be a need for risk mitigation through potential restrictions.<br><br>Regarding the evaluation of the monitoring data, the peer review concluded that the groundwater monitoring dataset for glyphosate and AMPA was insufficient to be directly used for regulatory exposure assessment without additional information (notably in terms of relation to use pattern of the active substance and temporal percentile). A thorough assessment against the quality criteria in the FOCUS report (European Commission, 2014; Sanco/13144/2010, version 3) would be required to select data that could be relevant for supplementing an assessment in the framework of Regulation (EC) No 1107/2009. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | relevant metabolites in groundwater in concentrations > 0.1 µg/L (even in the period 2009-2012 17).<br><br>*— LAWA, Report on the quality of groundwater – Plant protection product, 2019 (Annex A.23)*<br><br>Due to these data gaps and inconsistencies in the assessment, it cannot be excluded with the necessary certainty that glyphosate and/or its metabolites enter into groundwater at concentrations > 0.1 µg/L. | The publication mentioned in the document "*LAWA (2019): Bericht zur Grundwasserbeschaffenheit – flanzenschutzmittel – Berichtszeitraum 2013 bis 2016, Bund/Länder-Arbeitsgemeinschaft Wasser, 55p., Gotha."* was indeed considered during the peer review process and comprehensively evaluated together with all other groundwater monitoring data. |
| 8. | Failure to take account of effects on soil organisms<br><br>Page 38 (EN) [Page 43 (DE)] | **IV Individual analysis of the assessment failures and errors**<br><br>**6. Failure to take account of effects on soil/organisms**<br><br>The soils throughout Europe are heavily contaminated with glyphosate and its degradation products. Analysis of topsoils from 11 European countries and six different agricultural systems found glyphosate and/or AMPA in 45 % of samples.<br><br>*— Silva et al., Distribution of glyphosate and ami-nomethylphosphonic acid (AMPA) in agricultural topsoils of the European Union, 2017 (**Annex A.24**)*<br><br>A recent study carried out in France also shows that on arable landscape contamination with glyphosate is widespread in soil, but also in earthworms, suggesting that glyphosate is ingested by the soil biota. Glyphosate appears to have been detected in 88 % and AMPA in 58 % of soil samples, and 74 % and 38 % respectively | The risk assessment to soil organisms (i.e, earthworms, meso- and macro-fauna other than earthworms and soil microorganisms) was performed according to the current guidance document (European Commission, 2002. Guidance Document on Terrestrial Ecotoxicology Under Council Directive 91/414/EEC. SANCO/10329/2002-rev. 2 final, 17 October 2002). Regulatory studies conducted with all groups of relevant soil organisms with glyphosate, the formulation for representative uses and the relevant metabolite AMPA were used for the risk assessment while relevant and reliable peer-reviewed publications evaluating direct effects of glyphosate on soil organisms were not identified in the open literature in accordance with the criteria agreed at the Pesticides Peer Review Experts' TC 82. The soil organism risk assessment was completed using predicted environmental concentrations (PEC) calculated based on dissipation rates for glyphosate originating from field dissipation studies. Those for AMPA used information from laboratory incubations. It is true that a data gap was identified for AMPA dissipation rates from field experiments, but the risk characterisation for AMPA for the representative uses was concluded based on the laboratory decline information, which is an approach that is in line with the guidance in place. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | of earthworm samples.[74]<br><br>The available monitoring data for glyphosate and AMPA in soils were not sufficiently taken into consideration in the risk assessment (EFSA, Peer Review, p. 19 f.).<br><br>However, a comprehensive consideration of soil contamination and effects on soil organisms would have been necessary, especially in view of the moderate to very high persistence of AMPA in soil according to EFSA.<br><br>In addition, no reliable AMPA dissipation rates could be estimated from the available field studies, leading to the identification of a data gap (EFSA, Peer Review, pp. 17, 37) which is not acceptable for the renewal. | The publication mentioned in the document (Silva et al., 2018. Distribution of glyphosate and aminomethylphosphonic acid (AMPA) in agricultural topsoils of the European Union. Sci Tot Environ. 621:1352-1359) was considered in the peer review. The peer reviewed agreed that the measured concentrations of glyphosate and the metabolite AMPA from public monitoring programmes or literature articles for the soil compartment are only valid for the time and place they represent and are not equivalent to the predicted environmental concentrations in soil calculated for risk assessment purposes which resulted in higher concentrations so covered the monitored values. This is also the case for Pelosi et al. (2022) which clearly states that monitored soil concentrations were 10 times lower than predicted environmental concentrations. The findings reported in Pelosi et al. (2022) do not modify the exposure values that were used for the earthworm risk assessment for glyphosate and do not include any data that can be used for the hazard assessment. |
| 9. | Failure to take account of air spread<br><br>Air contamination<br><br>Pages 38-39 (EN)<br>[Pages 43-44 (DE)] | Indications of the presence of glyphosate and AMPA in ambient air and air particles were also not sufficiently taken into account.<br><br>Glyphosate is one of the most frequently detected substances when testing of pesticide concentrations in the air.<br><br>*— Kruse-Plaß et al, 2021 (**Annex A.25**)*<br><br>As a result, non-target species, including humans, may also be exposed to glyphosate residues via the ambient air. However, human exposure via inhaled air is apparently not sufficiently taken into account in the risk assessment.<br><br>*— Clausing, tree bark monitoring of pesticide pollution* | The peer review set some data requirements for the applicants regarding the collection of public monitoring data for the air compartment. The updated data collection, that included additional data from a French national exploratory pesticide campaign and information from monitoring studies in Germany (including Kruse-Plaß et al, 2021 that had results of Hofmann et al., 2019 "Biomonitoring der Pestizid-Belastung der Luft mittels Luftgüte-Rindenmonitoring und Multi-Analytik auf > 500 PSM-Wirkstoffe sowie Glyphosat". TIEM Integrierte Umweltüberwachung for: Bündnis für eine Enkeltaugliche Landwirtschaft e.V., Am See 1, 17440 Lassan; cited in Clausing (2020). http://www.tieminfo.de/.cm4all/uproc.php/0/Publikationen/Bericht-H18-Rinde-20190210-1518-1.pdf?_=16e5a98b3af&cdp=a.) and in France, was evaluated |

74 Pelosi et al., Glyphosate, AMPA and glufosinate in soils and earthworms in a French arable landscape, 2022, https://www.sciencedirect.com/science/article/abs/pii/S0045653522011651.

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | *over the Air: A toxicological assessment, 2020* ***(Annex A.26)***<br><br>These aspects do not appear to have been taken into account in the risk assessment on glyphosate. While EFSA considers it difficult to 'link the monitoring results of air samplers directly to the representative uses being assessed' (EFSA, Peer Review, p. 13), this does not alter their relevance for the assessment of whether the absence of unacceptable or harmful effects on the environment and health has been demonstrated. | and discussed extensively at the Pesticides Peer Review Experts' TC 81[75]. It was acknowledged (EFSA Conclusion, p. 20) that despite the few data available and the intrinsic properties of glyphosate, there was a high frequency of quantified samples with values >LOD (limit of detection) for glyphosate. However, the sampling apparatus (passive samplers) used in these studies measured particulate-bound glyphosate and not gas phase only. Transportation to air was therefore likely to be caused by wind-eroded particles transportation rather than volatilization or aerosols formed at the time of spraying. This route of entry is not specific to glyphosate - although it may be more apparent than for other substances due to the widespread use of the substance. However, it was highlighted that there is no specific regulatory framework in force in relation to this mode of transportation and a suggestion of future inclusion of data on particle-bound transport in the approval process should be taken into consideration in any future updates to the data requirements.<br><br>Regarding human health (as the Uniform Principles indicate that air concentrations are to be assessed against the human-toxicological threshold), the assessment for bystanders/residents was carried out with the EFSA model (belonging to the EFSA operator exposure guidance[76]). The default concentration in air was 1 microgram/$m^3$ (hence 0.001 mg/$m^3$), resulting in systemic exposure estimations for resident and bystanders below the (A)AOEL for all the representative uses proposed in the RAR. The risk characterisation is presented |

[75] See expert consultation point 4.6 in the Report of the Pesticides Peer Review Experts' TC 81 available in the Peer Review Report in the Open EFSA under 'Supporting documents' under EFSA Question number EFSA-Q-2020-00140: https://open.efsa.europa.eu/study-inventory/EFSA-Q-2020-00140 (refer to Part 3_Peer Review Report_Glyphosate_expert meeting reports_public.pdf (TC 81),

[76] EFSA (European Food Safety Authority), 2014. Guidance on the assessment of exposure of operators, workers, residents and bystanders in risk assessment for plant protection products. EFSA Journal 2014;12(10):3874, 55pp. https://doi.org/10.2903/j.efsa.2014.3874

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | | in the toxicology section. This default was above the monitored concentrations determined in air samples. |
| 10. | Failure to take into account effects on the microbiome<br><br>Page 39 (EN) [Page 44 (DE)] | **IV Individual analysis of the assessment failures and errors**<br><br>**8. Failure to take into account effects on the microbiome**<br><br>A drastic assessment gap is also that the potential effects of glyphosate on the microbiome identified in several studies have been77 completely ignored. This leads to an infringement of Article 4(3)(b) of Regulation (EC) No 1107/2009.<br><br>As regards the failure to take into account effects on microbiome, EFSA and the European Commission point out that there are currently no internationally agreed guidelines/harmonised criteria for the risk assessment of the microbiome in the field of pesticides and that further research is necessary to understand its relevance for the risk assessment and to develop strategies and methods (EFSA, Peer Review, pp. 3, 13).<br><br>However, the absence of guidelines is precisely the reason not to grant authorisation in view of lacking scientifically underpinned knowledge. However, the absence of guidelines can in no way be used as a justification for granting approval despite unclarified risks (see in this regard, C.II. and III. above). | It is disagreed that potential effects of glyphosate on the microbiome and possible consequence for health of humans, animals and the environment identified in several studies have been ignored.<br><br>On the contrary, an extensive investigation for such effects were made and the outcome of that assessments were reported in the conclusion. As regards non-target organisms, for example, some effects on bees' gut microbiota are clearly reported in the conclusion.<br><br>EFSA agrees that the lack of standardised guidelines is not a sufficient argument to dismiss possible effects on the gut microbiome. Nevertheless, in the absence of definitive information from the open literature, EFSA reiterates that the current assessment of glyphosate was based on a robust, up-to-standard data package and the derived current toxicological reference values for human heath are considered protective towards all the observed adverse effects, including those that could be secondary to gut microbiome perturbation, under the current state of knowledge.<br><br>As regards the identified effects on bees, it was considered that there was no ample evidence demonstrating that the health impact on honey bees has such importance which could have led to considerable colony level effects.<br><br>With regards specifically to the papers mentioned by Aurelia, Puigbo et al., 2022 reports about an in-silico study on potential targets for glyphosate on human microbiome. The authors |

77 Puigbo et al., Does Glyphosate Affect the Human Microbiota?2022, Life 2022, 12, 707, https://doi.org/10.3390/life12050707;
Barnett et al., Separating the Empirical Wheat From the Pseudoscientific Chaff: A Critical Review of the Literature Surrounding Glyphosate, Dysbiosis and Wheat-Sensitivity, 2020, Front. Microbiol. 11:556729, doi:10.3389/fmicb.2020.556729.

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | | admit that this necessitates support by empirical studies and epidemiological investigations to clarify the effect of glyphosate on the healthy human microbiome. Barnett et al., 2020, presents a literature review about possible relationship of glyphosate-induced microbiome dysbiosis and gastrointestinal diseases in humans, this since glyphosate would inhibit the shikimate pathway, a pathway exclusive to plants and bacteria. Various primary research studies on the microbiome are quoted and discussed, the large majority of these were duly assessed in the glyphosate peer review. Barnett et al reckon that the research surrounding glyphosate's effects on the gut microbiome suffers from numerous methodological weaknesses and call for future long-term studies examining physiologically relevant doses in both healthy and genetically susceptible populations to determine the real risk posed to human health. Overall, these two publications do not add evidence that change the current conclusions on glyphosate, and reinforce that additional work on microbiome is needed. |
| 11. | Incomplete assessment of clastogenicity (glyphosine)<br><br>Pages 39-40 (EN)<br>[Pages 44-45 (DE)] | In the absence of reliable results on the clastogenicity (i.e. the potential to cause DNA breaks) of glyphosate impurity glyphosine, EFSA identified a relevant data gap ('issue that could not be finalised') (EFSA, Peer Review, pp. 10, 31).<br><br>In view that no evidence of clastogenic potential was found in two in vivo tests and that the results of the positive in vitro studies were inconsistent, the Commission did not consider the data gap to be significant and set the maximum level of 3 g/kg for glyphosine, which it considered to be sufficiently protective (EU Commission, Glyphosate Renewal Report, p. 5).<br><br>On the basis of scientifically not validated assumptions, the European Commission overruled the gap identified | The impurity glyphosine showed a potential for clastogenicity in an *in vitro* chromosomal aberration assay that was not appropriately followed up *in vivo*; however, this impurity was present in some of the batches used in toxicity studies at levels representative of the proposed reference specification. Both relevance assessment and its maximum content was open for this impurity (not concluded, i.e. data gap), whereas there was evidence that it was present in some of the batches used in toxicity studies at levels representative of the proposed reference specification. Therefore this has led to an issue that could not be finalised.<br><br>It is acknowledged that Commission, together with MSs, in their role as risk manager, considered the impurity as toxicologically relevant and set a maximum level of 3 g/kg in the approval regulation during the decision-making phase**.** |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | by EFSA as relevant. In the applicants' view [of this review request], this approach is incompatible with the requirements of Regulation (EC) No 1107/2009, in particular with the precautionary principle. | |
| 12. | Incorrect assessment of the risk of cancer<br><br>Page 40 (EN) [Pages 45-46 (DE)] | *Paragraphs linked to classification and elements falling in the remit of ECHA*. | All pertinent studies on carcinogenicity were part of the hazard assessment undertaken in the context of the formal assessment of the proposal for harmonised classification and labelling in accordance with Regulation (EC) No 1272/2008 carried out by ECHA in parallel to the EFSA peer review, leading to the conclusions as delivered in the RAC Opinion on 30 May 2022 (ECHA, 2022).<br><br>The conclusions of EFSA result from the independent assessment of the data on carcinogenicity performed for the re-evaluation of glyphosate as a pesticide active substance. EFSA agrees on the conclusions reached in the ECHA RAC and by the assessment of the carcinogenicity studies and epidemiological data performed by the RMS in the RAR. |
| 13. | DNT findings<br><br>Page 41 (EN) [Page 46 (DE)] | **12. Other data gaps**<br><br>For example, a data gap has been identified to "determine whether the DNT findings reported in the studies with glyphosate trimesium and GBH are due to glyphosate" (EFSA, Peer Review, p. 37). The Renewal Regulation (EU) 2023/2660 does not address this point and does not provide for a condition of approval under Article 6 of Regulation (EC) No 1107/2009 in this regard. However, if a glyphosate salt at a dose considered safe for other glyphosate salts is found to be neurotoxic, it must be clarified without delay whether other glyphosate variants also exhibit this characteristic.<br><br>— *MIE/Ruden, What you don't know can still hurt you* | EFSA confirms that no DNT study was available in the dossier, however it was considered not needed based on the lack of evidence of concern for potential neurotoxicity in the dataset of regulatory studies on glyphosate active substance, including information on the chemical structure and pesticide mode of action of the active substance. During the risk assessment process, new evidence was brought forward, including public literature studies on glyphosate-based herbicides ('GBHs') in addition to the study on glyphosate-trimesium that was also made available to both the ECHA classification process and the EFSA peer review.<br><br>Following a weight-of-evidence assessment, EFSA concluded that there were no effects suggesting a DNT effect for |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | *– underreporting in EU pesticide regulation, 2022 (**Annex A.30**)*<br><br>The fact that the **toxicological reference values established ensure adequate protection against possible DNT effects** – according to the EFSA and the European Commission's argument regarding this data gap – **is not scientific evidence but a pure hypothesis.**<br><br>This data gap bears significant weight against the background that the legislator considers developmental neurotoxicity to be particularly serious (Annex II, No 3.6.1. Regulation (EC) 1107/2009, Annex Part A 5.6.2. of Implementing Regulation No 283/2013). | *glyphosate, while the same conclusion is not applicable for GBH or glyphosate-trimesium.*<br><br>A data gap was set by EFSA to cover the DNT uncertainties raised in the studies conducted with GBHs and with glyphosate-trimesium. The current toxicological reference values are considered however as sufficiently protective to cover these uncertainties and this was additionally substantiated by the literature study of Ojiro et al. 2023 where DNT relevant endpoints were not affected in the DNT sensitive population. |
| 14. | Incorrect handling of monitoring data<br><br>Page 41 (EN) [Page 47 (DE)] | Implementing Regulation (EU) 2023/2660 is also based on incorrect management of monitoring data.<br><br>The available data on concentrations of glyphosate and its metabolites in water, soil and air are often considered by EFSA to be irrelevant due to doubts as regards their representativeness (EFSA; Peer Review, pp. 13, 19 f.), although they provide indications of existing contamination and thus constitute an obstacle to approval.<br><br>In the view of the applicants' [of the current review process], this is inappropriate. Motoring data indicating risks must be taken into account in accordance with the precautionary principle.<br><br>In addition, the European Commission should have made provisions in the Renewal Regulation to ensure better monitoring of glyphosate and degradation products in soil, water and air. This task is also ineffectively "shifted" to the Member States. | An extensive review of existing monitoring data, including collection of public monitoring data and review of open literature, have been transparently evaluated by the peer review with regard to compliance of regulatory triggers and current guidance documents. From this comprehensive evaluation, it was clear that the results from these public surveys and literature reviews do not inherently fulfil the requirements of higher-tier assessments mandated by regulations such as Regulation 1107/2009. To utilise these data for regulatory purposes, additional information is necessary, and a comprehensive assessment against established quality criteria, such as those outlined in the FOCUS report (European Commission, 2014; Sanco/13144/2010, version 3), would be required. It was acknowledged that these results can still provide an overview of contamination levels in environmental compartments but it's essential to evaluate the context and reliability of the data before drawing conclusions or making regulatory decisions based on them. |

## Relevant scientific arguments provided in the review letter submitted by Secrets toxiques for the active substance glyphosate and the conclusions drawn by EFSA on the specific points raised

*It should be noted that the original request for internal review was provided in French language. Where available, a complimentary English translation has been provided to EFSA and ECHA by the Commission for the purpose to facilitate assessment by the Agencies. The English translation as displayed in column 2 has been generated by using an automated machine translation tool. Therefore the quality and accuracy of the translation may vary from the original text and should not be regarded as official translation. Only the original text of the request submitted in French should be considered as the authentic text.*

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| 1. | SECRETS TOXIQUES et autres / Requête en réexamen interne ré-approbation glyphosate - N/Réf. : 2400005/GT/AD<br><br>1. Background<br>Page 4 | - *The publication of the collective expertise of the National Institute of Health and Medical Research (INSERM) of 2021*[78] *"Pesticides and Health – new data" which confirms that "Studies on the exposure of the general population find widespread exposure to multiple pesticides"*[79]*, concludes inter alia that "As regards the herbicide glyphosate, the expert opinion concluded that there is an increased risk of NHL" (non-Hodgkin lymphoma) and calls for public action towards better protection of the population*[80]; | Epidemiological studies on possible association between exposure to glyphosate and incidence of non-Hodgkin's lymphoma (NHL) or other tumours were assessed in the peer review of the RAR and no conclusive evidence could be drawn that glyphosate exposure is associated with any cancer-related health effect. In specific relation to NHL, the EFSA Working Group on glyphosate noted inconsistencies as some meta-analysis found a modest association with glyphosate exposure (particularly the highest tertiles/quartiles) in the specific population groups of farmers and applicators occupationally exposed, most of them males. However, the most robust study (AHS agriculture cohort) did not find significant associations (Andreotti et al., 2018). In addition, the most recent study identified after public consultation (De Roos et al., 2022) found no association with the use of any herbicide, including glyphosate for an increased risk of NHL in a large, pooled study of 10 case-control studies from North America, the European Union and Australia. Nonetheless, a near-significant association was observed between glyphosate (lagged exposure 10 years) and follicular lymphoma, a subtype |

78 https://www.inserm.fr/wp-content/uploads/2021-07/inserm-expertisecollective-pesticides2021-rapportcomplet-0.pdf
79 see expert report p. 11
80 see pages XI and XII of the summary of the expert report.

| No. | Column 1<br>Reference to review letter | Column 2<br>Argument | Column 3<br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | | of NHL, although further research was considered to be needed. |
| 2. | 2. Background<br>Page 5 (EN)<br>[Page 6 (FR)] | As regards, more specifically, the impact of glyphosate-based formulations on the environment, the INRAE-IFREMER report on the impact of plant protection products on biodiversity and ecosystem services published on 14 September 2022[81] states that glyphosate pollutes all compartments (soil, water including marine, air). The following emerged:<br>'*Glyphosate and AMPA are among the PPPs with the highest detection frequency in European soils (Silva et al., 2019)' see p. 1367*.<br>As regards surface water contamination, a meta-analysis of 72 000 samples showed that "***glyphosate (…) and AMPA (…) were quantified in 43 % and 63 % respectively of the samples***" (see p. 1368). AMPA is the degradation by-product of glyphosate.<br>*"In ambient air, recent monitoring campaigns have also made it possible to quantify glyphosate in rural, urban and peri-urban areas with average detection frequencies close to 64 % and slightly increasing (to around 75 %) in arable crops, winegrowing and arboriculture areas".* | Regarding the soil compartment, the results for glyphosate and AMPA reported in the INRAE-IFREMER report and attributed to the Silva et al. (2019) publication, actually refer to the Silva et al. (2018) publication (refer to paragraph *2.2 Selection of the pesticide residues* in Silva et al., 2019). The publication Silva et al. (2018) was considered in the evaluation of the soil monitoring data. The peer review agreed that the measured concentrations of glyphosate and the metabolite AMPA from public monitoring programs or literature articles for the soil compartment are only valid for the time and place they represent and are not equivalent to the predicted environmental concentrations in soil calculated for risk assessment purposes which resulted in higher concentrations so covered the monitored values.<br>Regarding the surface water exposure, the meta-analysis cited in the INRAE-IFREMER report has been conducted by Carles et al. (2019)[82]. As indicated by the authors, the data on glyphosate and AMPA concentrations in surface waters in France were downloaded from the NAIADES public database. The same raw data from this database were included in the larger surface water public monitoring dataset evaluated by the peer review to address the aquatic exposure of glyphosate and AMPA. It was concluded that monitoring results from public surveys cannot be assimilated to concentrations that can be used for regulatory exposure assessment and be assessed |

81 https://hal.inrae.fr/hal-03777257/

82 Carles, L.; Gardon, H.; Joseph, L.; Sanchis, J.; Farre, M.; Artigas, J., 2019. Meta-analysis of glyphosate contamination in surface waters and dissipation by biofilms. Environment International, 124: 284-293

| No. | Column 1<br>Reference to review letter | Column 2<br>Argument | Column 3<br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | | against a regulatory exposure assessment goal without additional information (e.g. aspects such as agricultural context, including farmer usage of plant protection products, or site characterisation).<br><br>The aquatic risk assessment for glyphosate and AMPA adhered to the EFSA PPR Panel Guidance Document On Tiered Risk Assessment For Plant Protection Products For Aquatic Organisms In Edge-Of-Field Surface Waters In The Context Of Regulation (EC) No 1107/2009 (EFSA Journal 2013;11(7):3290, 268 pp. doi:10.2903/j.efsa.2013.3290), ensuring that the risk assessment process was conducted in accordance with established EU standards and methodologies specific to aquatic environments.<br><br>The peer review set some data requirements for the applicants regarding the collection of public monitoring data for the air compartment. The updated data collection, that included additional data from a French national exploratory pesticide campaign (including raw monitoring data for air downloaded from the public web database of the CNEP, from which the results reported in Column 2 are taken) and information from monitoring studies in Germany and in France, was evaluated and discussed extensively at the Pesticides Peer Review Experts' TC 81 [83] . It was acknowledged (EFSA Conclusion, p. 20) that despite the few data available and the intrinsic properties of glyphosate, there was a high frequency of quantified samples with values >LOD (limit of detection) for glyphosate. However, the sampling apparatus (passive samplers) used in these studies measured particulate-bound glyphosate and not |

[83] See expert consultation point 4.6 in the Report of the Pesticides Peer Review Experts' TC 81 available in the Peer Review Report in the Open EFSA under 'Supporting documents' under EFSA Question number EFSA-Q-2020-00140: https://open.efsa.europa.eu/study-inventory/EFSA-Q-2020-00140 (refer to Part 3_Peer Review Report_Glyphosate_expert meeting reports_public.pdf (TC 81),

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | | gas phase only. Transportation to air was therefore likely to be caused by wind-eroded particles transportation rather than volatilization or aerosols formed at the time of spraying. This route of entry is not specific to glyphosate - although it may be more apparent than for other substances due to the widespread use of the substance. However, it was highlighted that there is no specific regulatory framework in force in relation to this mode of transportation and a suggestion of future inclusion of data on particle-bound transport in the approval process should be taken into consideration in any future updates to the data requirements. |
| 3. | Failure to take into account existing studies on the representative formulation<br><br>Pages 25-26 (EN)<br>[Pages 27-29 (FR)] | A rapid review of the literature also identifies two other studies specifically on MON52276, highlighting alarming adverse effects. They are:<br><br>o Mesnage R, Teixeira M, Mandrioli D, Falcioni L, Ducarmon QR, Zwittink RD, Mazzacuva F, Caldwell A, Halket J, Amiel C, Panoff JM, Belpoggi F, Antoniou MN (2021). Use of shotgun Metagenomics and Metabolomics to Evaluate the Impact of Glyphosate or Roundup MON 52276 on the Gut Microbiota and Serum Metabolome of Sprague-Dawley Rats. Environ Health Perspect. 129: 17005. [84]<br><br>o Mesnage R, Ibragim M, Mandrioli D, Falcioni L, Tibaldi E, Belpoggi F, Brandsma I, Bourne E, Savage E, Mein CA, Antoniou MN. (2022) comparative Toxicogenomics of Glyphosate and Roundup Herbicides by Mammalian Stem Cell-Based Genotoxicity Assays and Molecular Profiling in Sprague-Dawley Rats. Toxicol Sci. 186: 83-101.[85] | Both cited studies were considered in the RAR. Mesnage et al. 2021 was considered by the EFSA Working Group on glyphosate in the context of the possible effects on microbiome and the following was concluded:<br><br>**Reliability**: Appropriate methodology to investigate bacterial populations was used. Some weaknesses and unclarities were identified: the study was neither conducted under GLP nor according to standardised regulatory test guidance; it is not clear how the sample size was defined. The composition of MON 52276 is not detailed.<br><br>**Relevance:** This in vivo study with associated adequate microbiological investigations provides some information of possible effects of glyphosate and MON 52276 on rat gut microbiome. It is noted that there are no guidelines for assessment of microbiome in the |

84 https://doi.org/10.1289/EHP6990
85 10.1093/toxsci/kcontraven143
https://pubmed.ncbi.nlm.nih.gov/34850229/

| No. | Column 1<br>Reference to review letter | Column 2<br>Argument | Column 3<br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | The first mentioned study (that of 2021) shows an **impact of both glyphosate and MON52276 on the intestinal microbiota** of rats, through the known mechanism of action of glyphosate on plants. It also detects **oxidative stress markers after exposure to MON52276 or glyphosate alone**. Finally, **the effects of MON52276 are generally more pronounced than those of glyphosate alone**.<br><br>The importance of intestinal microbiota for the preservation of health is essential[86].<br><br>INRAE explains its role: "*Since our birth, we have been living in symbiosis, in a win-win relationship, with the microbes that inhabit our body. In the intestine alone, we host as many bacteria as there are calls in our body! The proper functioning of this symbiosis is a health capital that we need to maintain. (...) For example, these micro-organisms: feed on nutrients that we cannot digest; protect us against environmental micro-organisms; continuously stimulate our natural immune defenses; interact with our human cells and tissues, locally with the intestinal wall but also distant from the liver or even the brain, producing in particular small molecules*."<br><br>A link between microbiota and certain diseases (cardiovascular diseases, autism) has been established. Oxidative stress is a type of attack against the constituents of the cell. It is a factor in inflammation and mutagenesis, but it is also considered, among others, one of the main causes of cancer.<br><br>This is not a long-term study but a 90-day study. | regulatory context. The relevance of this study is unclear.<br><br>**Conclusion**: Due to the above considerations this study does not allow to conclude on effects of glyphosate and MON 52276 on the gut microbiome and possible consequent impact on health, particularly as concerns metabolomics. The study is considered not to add elements to the current risk assessment of glyphosate.<br><br>Mesnage et al., 2022 was identified during the public consultation period and the RMS was requested to include it in the assessment for the overall weight of evidence related to the oxidative stress potential of glyphosate (cfr. Peer Review Report – Evaluation tables, Experts' consultation point 2.17[87]). |

86 https://www.inrae.fr/alimentation-sante-globale/microbiote_intestinal#:~:text=Depuis%20notre%20naissance%2C%20nous%20vivons%20en%20symbiose%20avec%20notre%20microbiote,nous%20garder%20en%20bonne%20sant%C3%A9

87 available in the Peer Review Report in the Open EFSA, section 'Supporting documents' under EFSA Question number EFSA-Q-2020-00140: https://open.efsa.europa.eu/study-inventory/EFSA-Q-2020-00140); refer to Part 4_Peer Review Report_Glyphosate_evaluation tables_public.pdf_(section 2)

| No. | Column 1<br>Reference to review letter | Column 2<br>Argument | Column 3<br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | **This study is not mentioned in the list of studies taken into account in the RAR.**<br>The second study (the 2022 study) also observed the presence of **oxidative stress in MON52276**. It also showed that, at equivalent doses of **glyphosate, MON52276 has greater effects of steatosis and hepatic necrosis than glyphosate alone**. Finally, genotoxic effects (oxidative stress and DNA degradation) were observed in kidney and liver cells. These effects are known to be a precursor to the development of cancers, particularly liver cancer.<br>This is not a long-term study but a 90-day study.<br>This study is cited in the list of studies taken into account in the RAR, but the adverse effects observed are not considered as such in the EFSA Conclusion.<br>**Both studies therefore show adverse effects of both MON52276 and glyphosate and should have been included in the assessment report.**<br>**There are therefore at least three studies showing adverse effects of MON52276. These studies provide experimental evidence of the toxicity of the representative formulation and provide evidence of its non-safety. However, they were not taken into account in the assessment report. No long-term studies were carried out**. | |
| 4. | **Failure to assess cocktail effects**<br>Exposure<br>Pages 26-27 (EN)<br>[Pages 29-30 (FR)] | **E- Failure to assess cocktails effects following the representative formulation and other treatments**<br>Cocktail effects were not evaluated because *"no combination of plant protection products is recommended on the label".*<br>Regulation (EC) No 1107/2009 provides that the assessment of compliance with its requirements and conditions of approval of a pesticide product '*under conditions of application* | In relation to the cumulative exposure to pesticide active substances, EFSA has been working to assess the cumulative risks of exposure to multiple substances. So far the dietary cumulative risk assessment has not shown exceedance of the risk thresholds for the target tissues/effects analysed (thyroid, neurotoxicity, chronic acetylcholinesterase inhibition and crano-facial malformations). Work is on-going to assess dietary |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | *consistent with good plant protection practice and realistic conditions of use' (cf. Article 4 (3)).*<br>The fact that the product label does not provide for simultaneous use with another formulation to carry out its herbicidal action does not correspond to assessment carried out under realistic conditions of use and environmental conditions. Cocktail effects are not limited to the effects of two commercial products intended to be applied simultaneously.<br><br>In addition to the diffuse pollution and persistence of certain products in the environment, in conventional agriculture, different treatments (herbicides, fungicides, insecticides) are applied to the same crop during the cropping cycle and the number of treatments may be particularly high, in particular in orchards or viticulture. Cultivated crops, farmers, residents and the environment are exposed to cocktails. Wine and orchard workers who regularly handle crops (crop monitoring, weeding, pruning, manual harvesting, etc.) are among the most highly exposed and have many cases of occupational diseases due to their multiexposure.<br><br>It should be noted that the exposure of agricultural workers did not take this multi-exposure into account. The assessment consisted of theoretical calculations, a priori, of exposure to the representative formulation. However, as their exposure estimates are considered theoretically lower than the toxicological reference value, **no real verification of the actual exposure of workers has been carried out**. | cumulative risks for other organs/effects (liver, kidney, reproductive and developmental effects) and to set up the methodology to perform non-dietary cumulative risk assessments. |
| **5.** | Insufficient assessment of indirect effects on biodiversity<br><br>INRAE report[88] | **F- Insufficient assessment of indirect effects on biodiversity**<br>(…)<br><br>It notes, in general, that 'biodiversity and ecosystem services | Specific data requirement and evaluation/decision making criteria for biodiversity and indirect effects are not available (as mentioned also in the INRAE- IFREMER report at page 1002). The need to develop a harmonised approach was also acknowledged during |

[88] https://hal.inrae.fr/hal-03777257/

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | Pages 27-30 (EN)<br>[Pages 30-34 (FR)] | are partially taken into account' (see p. 1001) and states that the regulatory agencies never assess the indirect effects on biodiversity:<br><br>'Indirect effects – cumulative – effects on biodiversity: In all cases, analysis of indirect effects (alteration of food resources) and more generally of trophic interactions in an ecosystem is not performed, although it is explicitly referred to in Regulation (EC) No 1107/2009.'<br><br>(….)<br><br>… INRAE Research Director, specialising in soil ecotoxicology, also criticised the tests carried out … used for assessment of long-term adverse effects on earthworms, the guarantors of soil fertility.<br><br>When assessing the effects of glyphosate-based herbicides, she considers that academic research has not been taken into account, but above all that regulatory tests are carried out over too short periods of time and using inappropriate (less sensitive) models to highlight long-term harmful effects on the environment and farming systems. This leads to an underestimation of the toxic effects of glyphosate based products.<br><br>The INRAE-IFREMER collective expert report notes 'the effects of glyphosate on microbiota (i.e. dysbiosis cases: disturbance of microbiota in terms of taxonomic and functional composition) were also observed in various terrestrial vertebrates including birds, mammals or amphibians (adult stage)." These effects also affect honeybee larvae. It notes that few studies are available on the effects on the diversity of aquatic invertebrates and on the effects on fish and amphibians (aquatic stage). | the peer review of glyphosate. By developing this approach, specific protection goals (SPGs) need first to be defined and agreed which would help to define specific data requirements and evaluation criteria.<br><br>Regarding the statement of INRAE Research Director, it is noted that risk assessment for soil organisms, as for other non-target organisms, follows a stepwise approach starting with worst-case assumptions in terms of exposure and using standard species tested in laboratory to predict the effects. When concerns are identified at lower tier, higher tier studies are requested to investigate the effects on abundance and biodiversity in field, under use condition of the plant protection product under evaluation. The scope of the risk assessment at all the tiers is to protect the soil ecological structure and, as a consequence, the soil function. It is noted that, with the definition of SPGs mentioned above, an advancement of the current approach could be envisaged in consideration of the specific level of protection given.<br><br>It is also noted that the protection of soil function, as of the biodiversity in general, should consider other factors, such as agricultural practices or landscape management, since it is a multifactorial issue as reflected in the EFSA Conclusion.<br><br>Academic studies were considered for the peer review of glyphosate, but relevant and reliable peer-reviewed publications evaluating direct effects of glyphosate on soil organisms were not identified in accordance with the criteria agreed at the Pesticides Peer Review Experts' TC 82.<br><br>Several studies from open literature on effects on microbiome of non-target organisms were considered |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | It points out: "*The case of glyphosate is emblematic as it highlights shortcomings in regulatory risk assessment (Robinson et al., 2020). (...) it showed that the legal framework determining the ecotoxicological risk assessment often leads to underestimation of risks and overestimation of the certainty and accuracy of assessments (Arcuri and Hendlin, 2019). The problem stems in particular from the use of standardised biological models. This was a crucial step to highlight the lack of a systemic approach to assessing the effects of PPPs on the environment, and the need for a holistic and inclusive approach for the assessment, not only based on life sciences and environmental data that fail to determine acceptable cumulative exposure and effects (Hamlyn, 2015; Leonelli, 2018).* '<br><br>(….)<br><br>The alleged lack of common methodologies or guidance does not exempt EFSA from carrying out a comprehensive assessment with the available data and methods, nor from having experimental tests carried out with the representative formulation to be requested by the industry applicant. | based on their evaluation for relevance and reliability according to the criteria agreed at the Pesticides Peer Review Experts' TC 82. As reported in the EFSA Conclusion, only for bees, the studies identified were evaluated as relevant and reliable and responses due to glyphosate exposure on bees' gut microbiota were identified, such as changes in the abundance of core microbial species. In particular, a decrease in abundance and growth of bee gut bacterium *Snodgrassella alvi* was observed. Generally, it was acknowledged that the relevance of these effects at the population level is unknown, e.g. it is difficult to link these effects to any impact on the colony strength.<br><br>EFSA, in its strategy, has already envisioned to advance the environmental risk assessment towards system-based approach and several projects[89,90] in the area of pesticides have been initiated to integrate landscape, ecology elements and evaluation of effects at higher level of biological organization.<br><br>The lack of common methodologies did not prevent EFSA from peer reviewing and extensively discussing the information provided at the Pesticides Peer Review Experts' TC 82. The outcome of this assessment is summarised in the EFSA Conclusion on page 24. The topic was also considered by the EFSA Working Group on glyphosate which delivered a position paper included as background document of the TC 82 report[91]. |

[89] https://www.efsa.europa.eu/en/events/webinar-ocefsaprev202302-call-tender-eu-environmental-scenarios-era-non-target-organisms
[90] https://www.efsa.europa.eu/it/art36grants/article36/euba-efsa-prev-2023-01-pera-advancing-era-plant-protection-products-towards
[91] available in the Peer Review Report in the Open EFSA under 'Supporting documents' under EFSA Question number EFSA-Q-2020-00140: https://open.efsa.europa.eu/study-inventory/EFSA-Q-2020-00140); refer to Part 3_Peer Review Report_Glyphosate_Annexes. TC 82.

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| 6. | Failure to take into account risks identified by studies of glyphosate-based herbicide formulations<br><br>Effects on other formulations<br><br>Pages 30-31 (EN) [Page 34 (FR)] | Regulation 1107/2009 provides that the approval of an active substance is to be carried out: "*in the light of current scientific and technical knowledge*" (cf. Article 4 (1))<br><br>Article 8 (5) provides that the applicant's dossier shall include:<br><br>"scientific peer-reviewed open literature, as determined by the Authority, validated by the scientific community and published in the last 10 years prior to the date of submission of the dossier, on side effects on health, the environment and non-target species of the active substance and its relevant metabolites."<br><br>However, the analysis of the RAR shows that relevant data have been discarded and some have not been included.<br><br>In addition to studies specific to MON52276, the RAR contains a number of studies including observations on other glyphosate-based herbicides, which tend to demonstrate often more toxic adverse effects of formulations compared to glyphosate alone.<br><br>However, these studies are at best considered as secondary, which is the case for most of the studies produced by international scientific literature outside the frameworks defined for studies produced by industry for product and substance approval purposes. Moreover, observations on formulations are systematically ignored on the grounds that the observed effects might not be due to glyphosate itself.<br><br>As a result of that distinction made by EFSA, a clear discrimination is visible between the consideration given to studies carried out by industry for the purposes of the approval procedure and the consideration given to studies carried out by the international scientific community for the purposes of their independent scientific research tasks. This differential treatment is contrary to the precautionary principle laid down in the TFEU (see CJEU judgment of 1 October 2019, | EFSA concurs that studies performed on products other than the representative formulation should not be disregarded a priori as non-relevant since they could potentially provide information as regards the toxicity of the active substance itself or information on potential higher toxicity of that formulation compared to the representative formulation. For this reason, the applicants were requested to disclose information on the composition of commercial PPPs to allow the assessment of the equivalence with the composition declared for the representative formulation and the interpretation of public literature toxicological and ecotoxicological studies conducted on GBHs**.**<br><br>The criteria followed in the peer-review process for the assessment of the relevance of the tested material and for the relevance and reliability of the endpoints were discussed in detail and agreed during the Pesticides Peer Review Experts' TC 82.<br><br>The request to applicants was addressed for only a number of the tested formulations while for others, the applicants did not provide a complete consideration of the composition of formulations used in the literature studies together with a consideration of whether the tested formulation was comparable to the formulation for representative uses, 'MON 52276'. The lack of such information may represent a source of uncertainty regarding the selection of the endpoints for risk assessment. Thus, a data gap was identified.<br><br>The available literature on GBHs that were available to the peer review were considered in the weight of evidence of all relevant endpoints. |

| No. | Column 1<br>Reference to review letter | Column 2<br>Argument | Column 3<br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | paragraphs 93 to 95), which requires equal weight to be given to studies from applicants and those from the international scientific community.<br>This failure to take into account of data on the effects of formulations has long been pointed out as one of the reasons for the divergent conclusions reached by institutions such as the International Agency for Research on Cancer (IARC) and the Institut National de la Santé et de la Recherche Médicale (Inserm), and the regulatory agencies. | |
| 7. | INSERM's collective expertise (meta-analysis)[92]<br><br>Pages 31 (EN)<br>[Pages 35 (FR)] | The collective expertise of the National Institute for Health and Medical Research (INSERM) of 2021 (see footnote 4) devoted an entire chapter to the human health effects of glyphosate and glyphosate-based formulations.<br>This expertise analysed studies from the international literature on glyphosate based formulations.<br>Unlike the RAR, it looked at epidemiological data stating that "*While regulatory agencies assess the toxicity to humans and the environment of active substances such as glyphosate, living organisms are not only exposed to them but also to co-formulants. This also applies to the vast majority of pesticides that are present in mixtures containing adjuvants of various kinds.*"<br>These studies show **significant increase in the risk of non-Hodgkin's lymphoma** (NHL) reinforcing IARC data from 2015, but also of **myeloma and some leukaemia** (with a lower level of evidence). They also show **neurodevelopmental disorders of children exposed prenatally, due to the proximity of farms within a radius of 500 m to 2 km around their residence.** | Epidemiological data were assessed as part of the peer review and it was concluded that the evidence for a causal association between the exposure to glyphosate and an increase in the incidence of NHL or other tumour types is not conclusive.<br>Similarly, no conclusive evidence could be drawn for the association with any neurological diseases or neurodevelopmental disorders.<br>Regarding genotoxicity, the weight of evidence (WoE) approach for genotoxicity on glyphosate active substance during the peer review included more than 70 studies (regulatory and public literature studies) both on the active substance and on GBHs. Studies were assessed as acceptable, supplementary or supportive (see Renewal Assessment Report at Table 2.0.5.2-3 and Table 2.0.5.2-4 (RAR, Volume 1, 2023)). Where available, the applicants provided information on the composition of the formulations (different from the representative one) used in published and non-published studies. Considerations on whether these formulations were comparable to the formulation for the representative uses were also included in the RAR |

[92] https://www.inserm.fr/wp-content/uploads/2021-07/inserm-expertisecollective-pesticides2021-rapportcomplet-0.pdf

| No. | Column 1<br>Reference to review letter | Column 2<br>Argument | Column 3<br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | As regards the genotoxicity of glyphosate, INSERM states:<br><br>"***studies showing a lack of genotoxicity of glyphosate appear to be less important both qualitatively or quantitatively than studies suggesting a positive effect***. *Whether a comparison is made between two rather similar studies showing opposite results, e.g. in mice (intraperitoneal exposure at 200 mg/kg glyphosate) (Rank et al., 1993; Mañas et al., 2009b), it is very interesting to note that the Mañas study incorporates a second injection 24 hours after the first, suggesting that repeated exposure (plausible phenomenon) is important to consider.* ***The timing of exposure to glyphosate or GBHs is therefore an important parameter. Moreover, the positive response is more pronounced with commercial formulations compared to the active ingredient when the studies are conducted in parallel (and thus comparable).***" (emphasis added)<br><br>*(see page 834 of the expert report accessible in footnote 4)* | Volume 4. In comparison with studies on the active substance, studies performed with formulations containing glyphosate, including the formulation for representative uses, were given a lower weight in the WoE for genotoxicity, due to the high uncertainties regarding potential different components of the formulations.<br><br>The studies performed with the formulation for the representative uses were considered in a WoE approach and it was concluded (EFSA, 2023)[93] that it is unlikely to be genotoxic or mutagenic. |
| 8. | INSERM's collective expertise (meta-analysis)[94]<br><br>Pages 32-33 (EN)<br>[Pages 36 – 37 (FR)] | For cytotoxicity studies:<br><br>**Of the 7 studies cited, 6 show a higher effect of** GBHs at much lower concentrations than sprayed solutions (during professional uses) that may come into contact with cells by oral route or more generally dermal.<br><br>For example, Mesnage [95] shows that the GBH can be 1 000 | Comments on pages 32 – 35 (EN) [36-39 (FR)] concerning potential higher toxicity of GBHs are acknowledged.<br><br>It should however be noted that EFSA's risk assessment processes are carried out in compliance with the relevant EU Regulations: the renewal process of glyphosate was conducted in accordance with the |

[93] EFSA (European Food Safety Authority),Alvarez, F., Arena, M., Auteri, D.,Binaglia, M., Castoldi, A. F., Chiusolo, A., Crivellente, F., Egsmose, M., Fait, G., Ferilli, F., Gouliarmou, V.,Nogareda, L. H., Ippolito, A., Istace, F., Jarrah, S., Kardassi, D., Kienzler, A., Lanzoni, A.,...Villamar-Bouza, L. (2023). Peer review of the pesticide risk assessment of the active substance glyphosate. EFSA Journal, 21(7),1–52, https://doi.org/10.2903/j.efsa.2023.8164

[94] https://www.inserm.fr/wp-content/uploads/2021-07/inserm-expertisecollective-pesticides2021-rapportcomplet-0.pdf

95 Mesnage R, Biserni M, Wozniak E, et al. Comparison of transcriptome responses to glyphosate, isoxaflutole, quizalofop-p-ethyl and mesotrione in the HepaRG cell line. Toxicol Rep 2018; 5: 819-26.

| No. | Column 1<br>Reference to review letter | Column 2<br>Argument | Column 3<br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | times more effective on the transcriptional response than glyphosate alone.<br>For mitotoxicity studies (mitochondrial toxicity):<br>Inserm cites three studies; two concern GBH alone**, the third is comparative and shows a stronger effect of the GBH.**<br>Concerning endocrine disruption: Inserm reviewed:<br>- in vitro studies on oestrogen-like effects and binding to the alpha androgenic receptor:<br>Four showed an effect of Glyphosate alone, 2 of the GBHs, and out of the 4 studies involving both glyphosate and the **GBHs two showed an effect at lower concentrations in GBH**. The fourth, Mesnage at al 2017, focused on the elucidation of a mechanism of action (activation of the alpha androgenic receptor) and not only on toxicity comparison.<br>- in-vivo studies:<br>Concerning developmental toxicity, the comparative study cited is in favour of an effect attributed to G; and 2 studies on GBHs show teratogenic effects.<br>With **regard to disturbance of reproductive functions (male and female), Inserm notes that the majority of these studies concern GBH:** 8 out of 15 focus on GBH alone, 6 are comparative GBH and G, and only one relates to G alone. The 3 comparative studies of the male reproductive system show comparable effects of G or GBHs. **On the other hand, the 3 studies on the female reproductive system showed a higher toxicity of formulations**.<br>These factors therefore make it possible to conclude that:<br>- epidemiological studies show effects of GBHs. It is therefore logical to study their toxicity. | standard procedures as laid down in Regulation (EC) No 1107/2009 and Commission Implementing Regulation (EU) No 844/2012.<br>Regulation (EC) No 1107/2009, that clearly defines the roles and responsibilities of EFSA and Member State competent authorities, stipulates that the assessment of active substances is carried out first at EU level as part of a peer review process with the Member States and EFSA based on the representative uses and formulation(s), whereas the assessment of other pesticide formulations is carried out at a second step by Member States, prior to authorisation of the formulations for use in their national territories, in accordance with the uniform principles as laid down in Commission Regulation (EU) No 546/2011.<br>Accordingly, the current regulatory process informs the conclusions drawn for herbicidal uses for the representative formulation of 'MON 52276' including its co-formulants. No conclusion has been made for uses with other formulations that may include other co-formulants/surfactants. Further authorised uses will be considered during the subsequent step, at product (re)authorisations where the "actual uses" of each PPP proposed for registration is evaluated at a zonal or national level, i.e. without an EU-wide scientific evaluation and involvement of EFSA in line with the dual system in place, by individual Member States before they give, or refuse authorisation for the use of pesticide formulations at national level. Such assessments duly consider the risks from the product in its entirety, taking into account the active substance(s), safeners, synergists and co-formulants it contains.<br>Nevertheless, EFSA also pays attention to potential toxicity that may arise from co-formulants and makes |

| No. | Column 1<br>Reference to review letter | Column 2<br>Argument | Column 3<br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | - the use of GBH studies is dominant in international literature;<br>- many toxicological studies (genotoxicity, cytotoxicity and even mitotoxicity) show a lower concentration effect of glyphosate used in GBH form.<br>- concerning endocrine disruption: as regards the study of in vitro mechanisms of action, the three comparative studies show twice a lower GBH concentration effect. In vivo, the situation is more complex: while the effects of G or GBH are comparable in males, studies on the female reproductive system show a greater toxicity of the formulations.<br>**This expert report therefore confirms that the international literature on the health effects of glyphosate-based herbicide formulations shows effects more adverse or different effects than those on glyphosate alone.**<br>Generations Futures carried out a comparison of data taken into account by Inserm and by the RAR on glyphosate, and it is very clear that EFSA excludes from the report data showing adverse effects, in particular in the long term, of glyphosate or its formulations, **by giving by default greater weight to industry studies than to scientific literature studies.** | necessary considerations in the peer review accordingly, as deemed appropriate. For this reason, studies performed on products other than the representative formulation were not disregarded a priori as non-relevant in the peer review as they could potentially provide information as regards the toxicity of the active substance itself or information on potential higher toxicity of that formulation compared to the representative formulation. The available literature on GBHs that were available to the peer review were considered in the weight of evidence assessment of all the relevant endpoints. |
| 9. | Higher toxicity of formulations / Exclusion of large number of data on other glyphosate formulations<br><br>Pages 34-35 [EN] | *Finally, in the particular case of a mixture = active substance + adjuvants, i.e. a commercial formulation,* ***several studies in particular on glyphosate have shown that the formulation has stronger effects than glyphosate alone*** *(Bianco et al. 2023; Mesnageet al.2015; Smith, Vera, and Bhandari 2019; Mesange et al. 2022). This was also underlined by the Inserm collective expertise of 2021 (Inserm 2021 collective expertise).*<br>*Thus, on the basis of all my work and international scientific* | See above. |

| No. | Column 1<br>Reference to review letter | Column 2<br>Argument | Column 3<br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | (Pages 38-39 [FR]) | *data, I confirm that the effects of chemical substances alone are not the same when they are found in a mixture. Therefore, mixtures should be studied as research objects "in their own right". In order to assess their potential harm, mixtures should be tested to assess their short and long term toxicity. This applies in the general case of mixtures of chemical substances and a fortiori, in the particular case where a mixture is composed of a pesticide active substance and several chemical adjuvants present to modify the properties of the active substance alone (commercial formulation), that is often difficult to dissolve or suspend and thus to pass through the cell barriers of the target organisms." (emphasis added)*<br><br>(…) said: *"I have carefully analysed the dossiers submitted by the applicants to request the re-authorisation of glyphosate and formulations (…)* ***Some studies are present in Volume 1 of the RAR for the evaluation of the active substance. The effects observed on the formulations are not included in Volume 3CP, which concerns the representative formulation****. I note 2 illustrative examples:*<br><br>- *Manservi et al 2019: the data discarded relate to in utero exposure to glyphosate and Roundup Bioflow for 13 weeks and show a negative impact on the development of the reproductive system and the hormonal regulation of pups.*<br><br>- *Pham et al. 2019: Discarded data also show that in utero exposure disrupts the regulation of sexual hormones in male pups. The authors also stressed the need to reduce the ADI of glyphosate for this observed harmful effect.*<br><br>*In addition, the list of exclusion criteria for publications (page 73 Glyphosate_RAR_01_Volume_1_2023-04-21_public) makes clear* ***that it should not take into account publications that concerned studies on glyphosate formulations other than the one considered*** | |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | ***representative for the assessment*** *(MON 52276 and Round Bioflow).* ***This criterion, in fact, excludes a large number of independent, peer-reviewed studies of high scientific value.*** *It is even more questionable given that there are a large number of glyphosate-based formulations that are widely used throughout the world and to which we may be exposed, and that any data alerting to certain harmful effects must be taken as a precaution for other formulations.*<br><br>*As a result, a large number of studies are excluded from the analysis and consideration of the results obtained (almost 70 based on PubMed data). This means ignoring a large number of toxic effects that are not considered in the acute and sub-acute toxicological studies provided by the applicants.* ***These dismissed effects*** *(non-exhaustive list)* ***concern cardiac toxicity, metabolic and hormonal deregulations, neurotoxic effects (autistic disorders), reproductive disorders, with effects on vulnerable periods of life (in utero exposure, birth, adolescence)****.*<br><br>*In the context of an informed re-authorisation of glyphosate, the best available knowledge should be taken into account, which is ultimately not the case here and with a view of minimising the harmful effects of glyphosate-based formulations on human health." (emphasis added)* | |
| 10. | Epidemiological data<br><br>Page 35 (EN)<br>[Page 39 (FR)] | **C-Epidemiological data**<br><br>**It is also noted that epidemiological data were not taken into account:**<br><br>*"epidemiological data, which relate precisely to the effects in humans of long-term exposure to glyphosate-based formulations, are not included in RAR-26 Volume 3 CP. For example, the meta-analysis of (Zhang et al. 2019) is not included in the meta-analysis, while authors report that results showing overincidence of certain blood cancers related to* | As regards epidemiological studies, all public literature submitted to EFSA throughout the regulatory process for the renewal of glyphosate (i.e. included in the RAR or requested during the public commenting phase) was considered as potentially relevant and included in the assessment. Additional literature identified after public consultation and considered appropriate was also included in the evaluation of epidemiological studies in the context of the peer review process. Public literature available to EFSA included primary research studies (case-control studies, cohort studies, etc.), narrative |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | *exposure to glyphosate formulations should be linked to experimental data from rodents (which are also discarded in RAR). This omission is unjustified and unjustifiable in terms of professional health. Another iconic and dismissed epidemiological study is the one carried out on the world's largest consortium of farmers (AGRICOH), which follows 3 million people, also shows overincidences of a certain type of blood cancer associated with exposure to formulated glyphosate (Leon et al. 2019).*<br><br>*Thus, with regulatory toxicological studies carried out only over short exposure periods and unjustified separation of experimental data in animals and epidemiological data in humans, the RAR's conclusions on the toxicity of formulations containing glyphosate ignore a large number of toxic effects in humans, in particular in workers and vulnerable populations. (…)* ***I conclude that the assessment of the long-term toxicity to humans of glyphosate-based herbicide formulations, and the renewal granted, do not seem to me to be neither relevant nor justified****".* | reviews, systematic reviews, meta-analysis, etc. To assess the available literature, EFSA with the support of the EFSA Working Group (WG) followed a structured approach. The scope of the activity of the WG was to assess the available information and provide a weight-of-evidence evaluation on the possible effects of glyphosate on human health. In addition, the WG included in its remit the evaluation of the RAR revised in September 2022. The outcome of such analysis can be found in Annex 4 of the Pesticides Peer Review Experts' TC 80[96]. Leon et al. 2019 and Zhang et al. 2019 are included in this analysis. |
| 11. | Scientific literature reviews **-** long-term adverse effects / exclusion of adverse effects from publications relating to other glyphosate-based products | **D- Scientific literature reviews**<br><br>The literature **reviews** on the long-term toxicity of glyphosate and its formulations all point to the existence **of long-term adverse effects**.<br><br>For example, the review published by Benbrook et al. (Benbrook, C. Mesnage, R. Sawyer, W. Genotoxicity Assays Published since 2016 Shed New Light on the Oncogenic Potential of Glyphosate-Based Herbicides. Agrochemicals | A robust assessment of all available data has been undertaken in the context of the EU peer review in an iterative process starting with the RMS assessment, followed by the peer review by EFSA and the Member States. This included also a rigorous evaluation of both industry studies submitted by the applicants and studies found in public literature, which were equally assessed for their relevance and reliability for the risk assessment |

[96] Available in the Peer Review Report in Open EFSA, Supporting documents section under EFSA-Q-2020-00140 (Part 3_Peer Review Report_Glyphosate_Annexes: Peer Review Report_Glyphosate_Annexes_TC 80_public.pdf)

| No. | Column 1<br>Reference to review letter | Column 2<br>Argument | Column 3<br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | Incorrect methodology by EFSA<br><br>Pages 35-36 (EN)<br>[Page 40 (FR)] | 2023, 2, 47 – 68.<br><br>https://doi.org/10.3390/agrochemicals2010005) analysing the results of studies on the genotoxicity of glyphosate and its formulations since 2016 – and thus since the publication of the IARC opinion – is categorical:<br><br>*"We assessed whether the tests published since the completion of the EPA and IARC reviews shed new light on the genotoxicity of glyphosate/GBH. We found 94 such tests, 33 testing technical glyphosate (73 % positive) and 61 GBHs (95 % positive). Seven out of seven in vivo human studies reported positive results. In* ***light of the genotoxicity results published since 2015, the conclusion that GBHs do not pose a risk of cancer through a genotoxic mechanism is untenable*** *" (emphasis added). GBH is the acronym for glyphosate based herbicide.*<br><br>Similarly, Rana et al. (Rana, I., Nguyen, P.K., Rigutto, G., Louie, A., Lee, J., Smith, M. T., & Zhang, L. (2023). Mapping the key characteristics of carcinogens for glyphosate and its formulations: A systematic review. Chemosphere, 139572) have shown that studies published since the IARC monograph have strengthened and expanded the evidence for carcinogenicity of glyphosate and its formulations:<br><br>*"In 2015, the IARC Working Group stated that it found strong evidence in favour of classification KC2 [genotoxicity] and KC5 [oxidative stress]; However, since then, many studies have been published, which not only provide additional data for the above-mentioned classification KC2 and KC5, but also provide robust evidence for KC4 [epigenetic effects], KC6 [Chronic inflammation] and KC8 [Alters receptor mediated effects]. There is also limited evidence in favour of KC1 classification [electrophilic chemical or can be metabolized to reactive electrophiles] and KC3 [impairs DNA repair or causes genomic instability]".* | and were taken into account in a weight of evidence approach**.**<br><br>To allow a transparent assessment of all the submitted studies, the RMS was asked to transparently report both the assessment of the reliability of the studies and the relevance of the study results to conclude on the overall weight of evidence.<br><br>Overall, all available studies, both from the applicants and from publications, have been duly considered and assessed for their relevance and reliability following a rigorous approach as detailed in the RAR. Where needed, additional information has been requested during the regulatory stop of the clock to complete the data package and address the comments received during the public and targeted MS consultation.<br><br>To facilitate consideration of studies with other formulations, where available, the applicants provided information on the composition of the formulations (different from the representative one) used in published and non-published studies. Considerations on whether these formulations were comparable to the formulation for the representative uses were also included in the RAR Volume 4. Depending on the availability of the evidence for the different toxicological endpoints, studies conducted with different salt-forms and/or formulations other than the representative one, were considered for their reliability and relevance and discussed as part of the weight of evidence in the risk assessment (see data requirement (general) 2.62 in Part 4_Peer review report_evaluation table (section 2)). Studies performed on products other than the representative formulation were not disregarded a priori as non-relevant in the peer review as they could potentially provide information as regards the toxicity of |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | The same review states that ***"studies of the highest quality indicate that glyphosate-based herbicides are genotoxic and have a greater effect than glyphosate alone",*** and that ***"exposure to glyphosate-based herbicide formulations causes endocrine disruption".***<br><br>The methodology used by EFSA to exclude publications relating to glyphosate-based products in their full composition or to give priority to industry studies is not consistent with the European regulation as interpreted by the judgment in BLAISE ruling of 1 October 2019. EFSA should use the same methodology as IARC or INSERM. | the active substance itself or information on potential higher toxicity of that formulation compared to the representative formulation. Therefore, available literature on GBHs that were available to the peer review were considered in the weight of evidence assessment of all the relevant endpoints.<br><br>In relation to the two cited publications: Benbrook et al (2023) was identified by the EFSA monitoring of new literature following the public consultation and it was screened for its impact on the risk assessment: the publication is a review and provided a list of genotoxicity assays published since 2016. After a screening of those publications published during 2018, 2019, 2020 included in the review, EFSA identified some publications not included in the assessment report (toxicology section). Most (except two) publications are related to genotoxicity studies performed on non-relevant species such as fish or reptiles; these publications are excluded from the genotoxicity assessment. A publication is a poster with very few details and it is unreliable for the genotoxicity assessment. The remaining publication is on adjuvant not approved in Europe and therefore not relevant for the current assessment. Overall, this review paper does not impact the overall weight of evidence for genotoxicity on glyphosate and the formulation for the representative uses as agreed during the peer review meeting.<br><br>The publication of Rana et al is dated October 2023 and has not been screened in view of its recent availability. |

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| 12. | Articles on human exposure to glyphosate /ADI<br><br>Pages 36-37 (EN)<br>[Pages 41 - 42 (FR)] | **E – Articles on human exposure to glyphosate**<br><br>As mentioned in the introduction, a wide-ranging campaign to analyse the exposure of French people to glyphosate was carried out, on the proposal of the glyphosate campaign association France, a founding member of the Secrets toxiques association. It involved 6848 screening and quantification analyses on urine samples carried out under the supervision by authorities. The results of this detection campaign have been submitted for statistical analysis. Refer to the study:<br><br>Grau D, Grau N, Gascuel Q, Paroissin C, Stratonovich C, Lairon D, Devault DA, Di Cristofaro J. Quantifiable urine glyphosate levels detected in 99% of the French population, with higher values in men, in younger people, and in farmers. Environ Sci Pollut Res Int. 2022 May;29(22):32882-32893. doi: 10.1007/s11356-021-18110-0. Epub 2022 Jan 12. PMID: 35018595; PMCID: PMC9072501.<br><br>The study can be found in footnote 7. It demonstrates: "***a general contamination of the French population, glyphosate being quantifiable in 99.8 % of urine samples with an average of 1.19 ng/ml +/– 0,84 after adjustment to body mass index (BMI). ".*** The study then investigated the links between the observed levels and various factors relating to the age, gender, occupation, living environment and lifestyle of the participants.<br><br>**This recent study, published in 2022 and carried out on a very broad scale, is not part of the body of studies of the RAR evaluation**.<br><br>However, it confirms the widespread exposure of French people to glyphosate, even though the vast majority of the participants tested are not users of glyphosate-based herbicides. Such exposure data should therefore generate attention of public authorities, first and foremost health | The study by Grau D et al, 2022, has been considered during the peer review in the context of biomonitoring data.<br><br>The main drawback of this study was the limited reliability of the analytical method/protocol (Elisa method) used for the urine samples analysis.<br><br>Urine samples were analysed according to the manufacturer's protocol, as validated by Krüger et al. (Krüger et al, 2014 also assessed during the peer review) based on ELISA and GC–MS assay data comparison on human urine samples. However, based on the study by Krüger et al, it is clear the Elisa protocol used was not demonstrated to be reliable for analysis of glyphosate in urine samples.<br><br>The study by Grau et al, 2023, is referring mainly to Zoller et al, 2020, and Niemann et al, 2015, both assessed during the peer review.<br><br>Zoller et al, 2020 was considered reliable with restrictions during the peer review, with human data based on a relatively low sample size (n=12) and suggesting an oral absorption value of 1% based on urinary excretion. Due to these limitations, the experts agreed that an oral absorption value of 20% is still valid for the risk assessment.<br><br>Notwithstanding this conclusion, in Annex 10 of the Pesticides Peer Review Experts' Meeting TC 80, both oral absorption values were used to estimate the systemic exposure levels based on biomonitoring data (urinary concentrations). For all EU data, both oral absorption values (1 and 20%) did not lead to an exceedance of the ADI or ARfD. |

| No. | Column 1<br>Reference to review letter | Column 2<br>Argument | Column 3<br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | agencies and competent authorities.<br><br>A second recent article on how to estimate human exposure has not been taken into account. This is a publication from 2023:<br><br>Grau D, Grau N, Paroissin C, Gascuel Q, Di Cristofaro J. Underestimation of glyphosate intake by the methods currently used by regulatory agencies. Environ Sci Pollut Res Int. 2023 Sep;30(45):100626-100637. doi: 10.1007/s11356-023-29463-z. Epub 2023 Aug 28. PMID: 37639106.<br><br>That article demonstrates that **the formula commonly used (Nieman's formula) by agencies to calculate, from the concentration of urinary glyphosate detected in a sample, the theoretical dose of glyphosate ingested by a human largely underestimates the actual dose ingested by that human**. **The article shows an average underestimation of around 34 times and over 53 times for half of the observations!**<br><br>It states *"our objectives were to test the robustness of the mathematical model currently used to calculate the daily intake of glyphosate, and to propose alternative models based on urinary excretion kinetics.* ***Our results show that the amount of glyphosate ingested is systematically underestimated by the model currently used by safety agencies, while the other models studied show better estimates****, with gender differences. Our results also show a high degree of inter-individual variability, leading to uncertainties particularly with regard to the link with the ADI, and further confirm that glyphosate excretion varies considerably between individuals following a similar dosing regimen.* ***In conclusion, our study highlights the unreliability of the assessment processes carried out by safety agencies, in particular for glyphosate, and pesticides in general, and questions the relevance of*** | |

| No. | Column 1<br>Reference to review letter | Column 2<br>Argument | Column 3<br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | ***such processes which are supposed to safeguard human health and the environment."*** | |
| 13. | ADI and biomonitoring (oral absorption and calculation of external exposure)<br><br>Pages 37-38 [EN]<br>[Pages 42-43 [FR]) | This ADI is calculated for the active substance glyphosate alone on the basis of animal experiments (NOAEL determination – No Observed Adverse Effect Level). An empirical safety factor of 100 is applied to convert this dose from the animal to humans and to take into account the variabilities of sensitivity within the human species.<br><br>However, we have seen above that:<br><br>- On the one hand, glyphosate-based herbicide formulations that are placed on the market and applied in the environment have a higher toxicity than glyphosate alone (up to 10 000 times for cytotoxicity observed in *in vitro testing*); and<br><br>- On the other hand, from life *in utero* onwards and throughout our lives, we are exposed to a number of chemicals that pollute our environment and are therefore exposed to a cocktail.<br><br>Thus, the factor 100 chosen as the safety factor for calculating the ADI seems in fact relatively unprotective.<br><br>In order to assess the risk arising from the ingestion of glyphosate, the safety agencies compare the daily intake of glyphosate with the ADI. As the formula used is flawed or at least disconnected from the reality in the field, its use for risk management results in a biased assessment of the actual exposure. The conclusions on the risks associated with this chronic exposure are therefore questionable.<br><br>The authors of the article state in particular that:<br><br>- the reference study for the establishment of the ADI during the re-approval process does not cover sufficiently long time period to be used for this purpose (a 90-day dog | The uncertainty factor of 100 is the standard factor applied for the derivation of the ADI, taking into account intraspecies and interspecies variability. Deviations from this standard factor to higher cumulative uncertainty factors are warranted if specifically justified. However, no need to increase the uncertainty factor was identified from the assessment of the available data.<br><br>The NOAELs selected for the derivation of the chronic health-based guidance values are lower than the NOAELs identified in long-term and reproductive toxicity studies and therefore provide sufficient protection for possible long-term effects of glyphosate.<br><br>In relation to comments on the specific toxicity of GBHs and on the possible cocktail effects, reference is made to responses provided in previous comments.<br><br>The recent human data were considered to present limitations, leading to the conclusion by the experts to maintain an oral absorption value of 20% for the risk assessment, as previously indicated. Nevertheless, it is reiterated that both oral absorption values (20% and 1%) were used to estimate the systemic exposure levels based on biomonitoring data (urinary concentrations). For all EU data, both oral absorption values did not lead to an exceedance of the ADI or ARfD. |

| No. | Column 1<br>Reference to review letter | Column 2<br>Argument | Column 3<br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | study, i.e. a study that does not cover the entire life of a dog);<br>- whereas Canada has set a lower ADI than Europe (0.3 mg/kg bw/day in Canada versus 0.5 mg/kg bw/day) based on a 2-year (i.e. lifetime) rat study,<br>- EFSA uses for its exposure calculations an outdated oral absorption rate of glyphosate (derived from old animal data), whereas recent human data are available and the German RMS representative suggested to take them into account. The use of this outdated oral absorption rate ***"leads to a risk 20 times higher than claimed by the agencies"***<br>- the results vary considerably depending on the time of sampling.<br>They conclude by explaining their work:<br>*'Niemann's formula using a urinary excretion rate of 20 %, instead of 1 % as recommended by Zoller and Faniland, leads to a mechanical underestimation by a factor of 20 of the quantity ingested and thus of the risk involved. However, this method of estimating the ingested quantity does not take into account the wide variations between individuals, depending on their age, gender and the time of sampling, which may lead to a much greater underestimation, thus significantly minimising the risks incurred by the population. Currently, no scientific study can assess the risk of permanent exposure to glyphosate for almost the entire population, as shown by Grau et al.*<br>*CONCLUSION*<br>*As it is currently designed, the assessment of the long-term toxicity of glyphosate to humans involves 3 steps, the determination of the ADI, the determination of oral absorption, and the link between the quantity in the urine and the quantity* | |

| No. | Column 1<br>Reference to review letter | Column 2<br>Argument | Column 3<br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | *ingested.*<br>*The determination of the ADI is questionable, the determination of oral absorption is erroneous, the formula used to determine the link between the quantity in the urine and the quantity ingested is unreliable. Thus, whatever the value of the ADI, the risk to the population remains unknown.'* | |
| 14. | ADI vs long-term exposure<br><br>Page 39 [EN]<br>(Page 44 [FR]) | Regarding the exposure of the general population, INSERM also questioned the protective value of the ADI in a context of repeated long-term exposure:<br>*"The quantification of glyphosate in urine is the most appropriate method for estimating and monitoring population exposure over time. (...) Urinary concentrations frequently found in occupationally exposed populations or in the general population are in the order of µg/l. These values are lower by a factor of 100 to 1 000 than those expected for chronic exposure corresponding to the ADI currently determined by EFSA, i.e. 0.5 mg/kg/d (EFSA, 2015a).* ***However, this reference value, based on experimental data in laboratory animals, does not exclude any risk to humans, in particular in case of repeated and long-term exposures****."*<br>*See p. 853 INSERM 2021 report accessible in footnote 4.*<br>In short, beyond the context of the RAR on glyphosate, the scientific literature, whether produced by research laboratories or generated by public research institutions in meta-analyses, indicates long-term adverse effects on carcinogenicity, reprotoxicity, neurotoxicity and endocrine disruption with a high degree of certainty, for both glyphosate and herbicides containing it. | Conversely, from the conclusion of INSERM, no clear evidence of causal association between exposure to glyphosate and onset of the cited pathologies was concluded from the assessment of the available epidemiological studies in the peer review. As noted by INSERM, the urinary levels would suggest a relatively low exposure, although there are uncertainties in the method to calculate exposure levels from biomonitoring data on glyphosate, as previously discussed. |
| 15. | Section 3.4<br>Implementing Regulation (EU) | Implementing Regulation (EU) 2023/2660 violates the right to live in a healthy environment<br>The elements of human exposure and contamination of the | The ECHA RAC Committee concluded in 2022 that no classification of glyphosate is warranted for adverse effects on reproduction and development. |

| No. | Column 1<br>Reference to review letter | Column 2<br>Argument | Column 3<br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | 2023/2660 violates the right to live in a healthy environment<br><br>Page 42 [EN]<br>(Page 48 [FR]) | environment and of the food chain presented above raise the issue of health and environmental risks in the first place. However, when this pollution reaches the blood of the umbilical cord and the meconium of newborns, it also raises the question on the acceptability of the situation where the mere evidence of widespread contamination is equivalent to a failure to protect the environment and populations. | Additionally, the peer review concluded that there was no clear pattern of effects suggesting a DNT effect for *glyphosate,* and the current toxicological reference values were considered as sufficiently protective (see also Pesticide Peer Review TC 80, Expert consultation 2.27[97]).<br><br>Finally, when considering the available epidemiological studies, no conclusion could be drawn on any potential causal association between glyphosate exposure and reproductive endpoints (see also Pesticide Peer Review TC 80, Expert consultation 2.7 identified following comments by public).<br><br>From abstract screening, the cited study (Kongtip et al., 2017) reports the serum levels of glyphosate in Thailandese pregnant women and in the umbilical cord at childbirth, showing that in subjects where glyphosate was detected, levels in mothers' serum were significantly higher than those in foetal serum (median: 17.5, range: 0.2-189.1 ng/mL vs median: 0.2, range: 0.2-94.9 ng/mL). No information is available on the exposure conditions, however it is noted that higher levels are measured in women working in agriculture, living in agricultural areas or having a family member working in agriculture. A literature screening identified another paper measuring maternal and foetal serum glyphosate levels in a group of 30 pregnant and 39 non-pregnant women living in a Canadian urban area and with no occupational exposure or in contact with occupationally exposed subjects. Glyphosate was not detected in serum of pregnant women or in the umbilical cord. Non-pregnant women had a glyphosate mean level of 73.6 ± 28.2 ng/ml (Aris and Leblanc |

[97] Available in the Peer Review Report in Open EFSA, Supporting documents section under EFSA-Q-2020-00140 (Part 3_Peer Review Report_Glyphosate_expert meeting reports_public (TC 80)

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| | | | 2011[98], reviewed in Gillezeau et al 2019[99]). Overall, an unclear picture emerges from these data; nevertheless, the scarce available evidence would not indicate a widespread contamination but rather localised hotspots possibly related to different dietary habits and lifestyles, specific conditions of use of pesticides and/or agricultural practices. |

[98] https://www.sciencedirect.com/science/article/pii/S0890623811000566
[99] https://d-nb.info/1178543129/34

## Relevant scientific arguments provided in the review letter submitted by Antidote for the active substance glyphosate and the conclusions drawn by EFSA on the specific points raised

*It should be noted that the original request for internal review was provided in French language. Where available, a complimentary English translation has been provided to EFSA and ECHA by the Commission for the purpose to facilitate assessment by the Agencies. The English translation as displayed in column 2 has been generated by using an automated machine translation tool. Therefore the quality and accuracy of the translation may vary from the original text and should not be regarded as official translation. Only the original text of the request submitted in French should be considered as the authentic text.*

| No. | Column 1<br><br>Reference to review letter | Column 2<br><br>Argument | Column 3<br><br>EFSA's scientific views on the specific point |
|---|---|---|---|
| 1. | DEMANDE DE RÉEXAMEN INTERNE [Article 10 du Règlement n 1367/2006 pris en application de la Convention d'Aarhus]<br><br>II.3. Infringement of the provisions of Regulation (EC) No 1107/2009 (i) Page 8/10 | For example, two studies published in the National Library of Medicine in June 2019 and December 2019 highlight the genotoxicity of glyphosate and glyphosate-based herbicides in human peripheral white blood cells and the toxicity of co-formulants when mixed with glyphosate[100]. | The cited study of Nagy et al. (2019) was included in the weight of evidence for the genotoxicity assessment performed in the RAR and in the preliminary assessment conducted by the EFSA Working Group on glyphosate. The study showed negative results for glyphosate, whereas equivocal results were observed when various glyphosate-based herbicides (GBHs) were tested, showing DNA damage in the presence of cytotoxicity. The study was evaluated as of limited reliability due to a series of methodological limitations.<br><br>The cited publication of Mesnage et al. (2019) is a review paper describing narratively the chemical identification and toxicity profile of some co-formulants (notably including co-formulants currently not allowed for use in PPPs in the EU (Polyethoxylated tallowamine surfactants), and their replacements. No original data is presented. The assessment of the toxicological profile of the formulation for representative uses was included in the RAR. |

100https://pubmed.ncbi.nlm.nih.gov/30951798/ https://pubmed.ncbi.nlm.nih.gov/31678731/