Pre-submission Meeting with AGG (RMSs: the Netherlands, Hungary, France & Sweden)

Glyphosate EU Annex I Renewal - Ecotoxicology

October 17, 2019

Glyphosate Task Force Ecotoxicology Technical working Group

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Proposed Glyphosate Uses for Annex I Renewal

[Please be aware that the GAP tables are still being developed and further uses may appear on the final GAP table after due consideration by the GTF2 member companies.]

Pre-sowing and pre-planting use in annual crops [Renovation / change of land use]

Crop and/or situation (a)	MS Country	Product name	F G or I (b)	Pests or Group of pests controlled (c)	Formulation		Application				Application rate per treatment			PHI (days) (m)	Remarks
					Type (d-f)	Conc. a.s. (i)	method kind (f-h)	Growth Stages & season (j)	number min-max (k)	Interval between application min-max	L/ha product min-max (l)	Water L/ha min-max	kg a.s./ha min-max (l)		
Root & tuber vegetables Bulb vegetables Fruiting vegetables Brassica Leafy vegetables Stem vegetables Sugar beet	EU	MON 52276	F	*Emerged weeds*	SL	360 g/L	Spray	Pre-sowing, crop pre-emergence / pre-planting, pre-transplanting.	1-3	21	2	100-400	0.72	-	Renovation / change of land use applications. Broadcast tractor mounted application (to 100% of the field). Use 75% drift reducing nozzles. Maximum application rate of 2.16 kg as/ha glyphosate in any 12 months period across all use types.

Pre-sowing and pre-planting use in annual crops [Post-harvest use]

Crop and/or situation (a)	MS Country	Product name	F G or I (b)	Pests or Group of pests controlled (c)	Formulation		Application				Application rate per treatment			PHI (days) (m)	Remarks
					Type (d-f)	Conc. a.s. (i)	method kind (f-h)	Growth Stages & season (j)	number min-max (k)	Interval between application min-max	L/ha product min-max (l)	Water L/ha min-max	kg a.s./ha min-max (l)		
Root & tuber vegetables Bulb vegetables Fruiting vegetables Brassica Leafy vegetables Stem vegetables Sugar beet	EU	MON 52276	F	*Emerged weeds*	SL	360 g/L	Spray	Post-harvest, pre-sowing / pre-planting.	1-3	21	2-6	100-400	0.72-*2.16	-	Application to existing row cropland after harvest for removal of remaining crop / stubble and for control of actively growing annual weed. Broadcast tractor mounted application (to 100% of the field). Use 75% drift reducing nozzles. Maximum application rate of 2.16 kg as/ha glyphosate in any 12 months period across all use types.

**For bulb and root vegetables (not tuber vegetables or sugar beet) a slightly lower maximum application rate (1.44 kg/ha) may be necessary, as residues per unit dose (RUD) value used in mammal chronic assessment is 48.3 and not 21.7, So, for BBCH ≥ 40 & 50, being more explicit in the comments of the GAP table indicating 'ground directed application for control of actively growing annual weeds after BBCH ≥ 40 and ≥ 50 including post harvest removal of crop remains / stubble.*

Use in top fruit plantations and vineyards

Crop and/or situation (a)	MS Country	Product name	F G or I (b)	Pests or Group of pests controlled (c)	Formulation		Application				Application rate per treatment			PHI (days) (m)	Remarks
					Type (d-f)	Conc. a.s. (i)	method kind (f-h)	Growth Stages & season (j)	number min-max (k)	Interval between application min-max	L/ha product min-max (l)	Water L/ha min-max	kg a.s./ha min-max (l)		
Top fruit plantations (including citrus, tree nuts, pome fruit, stone fruit, kiwi and banana)	EU	MON 52276	F	*Emerged weeds*	SL	360 g/L	Spray	Post-emergence of weeds	1-3	21 days	2-4	100-400	0.72-1.44	7	Avoid crop contamination during treatment. Maximum application rate of 2.88 kg as/ha glyphosate in any 12 months period Applications are performed in the rows below the trees or as spot treatments. The rate refers to the treated area only which represents not more than 50% of the total orchard area. The application rate with reference to the total orchard or vineyard surface area is not more than 50% of the stated dose rate.
Grape plantations (table and wine grape)	EU	MON 52276	F	*Emerged weeds*	SL	360 g/L	Spray	Post-emergence of weeds	1-3	21 days	2-4	100-400	0.72-1.44	7	

Status of Ecotoxicological risk assessment for glyphosate considering proposed GAP.

Considers max proposed GAP table rate

Expected outcome of Ecotoxicology risk assessment – assumes max single application rate of 2.16 kg/ha

Avian acute and chronic risk	Acute & chronic bird risk assessment - Pass at Tier II after refinement
Mammal acute and chronic risk	*Acute & chronic mammal risk assessment - Pass at Tier II after refinement
Fish acute and chronic risk	Acute & Chronic PASS at FOCUS Step 2 or 3
Aquatic Invertebrate acute and chronic risk	Acute & Chronic - PASS at FOCUS Step 2
Pollinator Risk Assessment	Risk assessment PASS at first Tier
Arthropods other than bees	Risk assessment PASS based on extended study data
Aquatic plants algae and macrophytes	Acute & Chronic PASS at FOCUS Step 2
Soil organisms incl. soil microbes	Chronic risk assessment PASS at first Tier
Non-Target Terrestrial Plants (NTTPs)	Risk assessment PASS with limited mitigation

*Consider 1.44 kg/ha for root and bulb crops BBCH ≥ 40 & 50

Status of Ecotoxicological risk assessment for glyphosate considering proposed GAP.

'Relevant and reliable endpoints from the public literature will also be considered in the risk assessment.'

Max single rate = 2.16 kg/ha.

(Endpoints for technical and formulation studies presented in terms of glyphosate acid)

PECsoil max = 3.84 mg/kg soil dw

PECsw (worst case); FOCUS Step 1 = 491 µg/L
Step 2 N & S EU = 234.7 µg/L
Step 3 global max = 11.102 µg/L

[Risk assessment for metabolites falls within that of parent, therefore only gly exposure data presented here]

Avian acute and chronic risk assessment	Expected RA outcome;

// Acute oral LD_{50} 4334 mg/kg bw;
Reproductive NOAEL = 96.3 mg/kg bw/d.
// Refinements needed; Yes - foliar residue decline – herbivorous birds
	Acute: Pass at Tier I
	Chronic: Pass at Tier II after refinement

Mammal acute and chronic risk assessment	Expected RA outcome;

// Acute oral **LD_{50} > 2000 mg/kg bw**
// Reproduction **NOAEL = 50 mg/kg bw/d.**
// Chronic endpoint relevance to wild mammal assessment
// Residue decline on dietary items (grass & BLW decline)
// Higher tier semi-field effects (enclosure study supporting orchards)
// Population modelling / Weight of evidence
	Acute mammal risk assessment
	(geomean acute endpoint applied to assessment)
	- Pass expected.
	Chronic mammal risk assessment
	- Pass at Tier II after refinement

Fish acute and chronic risk assessment	Expected RA outcome;

// Acute **LC_{50} 38 mg/L**; AMPA **LC_{50} = 520 mg/L**
// MON 52276 **LC_{50} >277& > 306 mg/L**
// Chronic NOEC = 1000 µg/L (*Brachydanio rerio*)
// Study not considered valid - Not most acutely sensitive species
// Env. concerns / test design / val. criteria / chem analysis
// Most relevant long term fish NOEC = 9.6 mg/L (trout ELS)
	Acute & Chronic
	PASS at FOCUS Step 2
	(FOCUS STEP 3 based on 1000 µg/L)

MON 52276 = representative formulation for the Annex I renewal

Aquatic Invertebrate acute and chronic risk assessment	Expected RA outcome;

// Acute EC_{50} 40 mg/L
// AMPA EC_{50} = 690 mg/L; HMPA EC_{50} > 100 mg/L
// MON 52276 EC_{50} = 209 mg/L
// Chronic;
// NOEC = 12.5 mg/L
// NOEC > 1000 mg/L- ***new study***
	Acute & Chronic risk assessment
	- Anticipate PASS at FOCUS Step 2

PECsw (worst case); FOCUS Step 1 = 491 µg/L
Step 2 N & S EU = 234.7 µg/L

Pollinator Risk Assessment	Expected RA outcome;

// **Honey-bee** acute contact & oral
// Glyphosate: LD_{50} >100 & 100 µg/bee
// MON 52276: > 100 & > 77 µg/bee
// Chronic NOAEL = 301 mg/L (colony feeding study)
 	
// **Non-*Apis sp.***;
// Acute solitary bee: LD_{50} > 461 µg /L
// Acute bumble bee oral & contact: LD_{50} > 412 & > 461 µg/L
// Chronic bee study (10 day lab adult feeding study);
LDD_{50} >179.9 µg /L
NOED >10000 mg/kg diet
// 22 Day bee larval toxicity; NOEDD > 500 ppm (*expected*)
	Pass at First Tier
	- Supported by colony feeding study, chronic lab and non-apis studies

Arthropods other than bees	Expected RA outcome
// *Extended lab studies with MON 52276 (representative formulation)* // *A. rhopalosiphi;* Ext. lab LR_{50} >5760 g/ha (>16 L MON 52276/ha) // *T. pyri;* Ext. lab LR_{50} ≥4320 g/ha (≥12 L MON 52276/ha) // *A. bilineata;* Ext. lab LR_{50} ≥4320 g/ha (≥12 L MON 52276/ha) // Chronic effects; (reproduction (R), fecundity (F) & parasitisation efficiency (PE); // *A. rhopalosiphi;* PE vs. control < 35% effects at 2.16 kg/ha // *T. pyri;* F vs. control < 25% effects at 2.16 kg/ha // *A. bilineata;* R vs. control < 20% effects at 2.16 kg/ha	**- PASS expected based on extended study results** **- In-field & Off-field HQs << 2 at max rate <2.16 kg/ha.** **- sub-lethal effects <35% at proposed max rate**

Aquatic plants algae and macrophytes			Expected outcome
Algae			
Anabaena flos-aquae;	$EC_{50(biomass / rate)}$	= 8.5 / 22 mg/L	
MON 52276	$EC_{50(biomass / rate)}$	= 55 / 88 mg/L	
AMPA	$EC_{50(biomass / rate)}$	= 110 / 200 mg/L	
HMPA	$EC_{50(rate \& biomass)}$	> 115 mg/L	
Macrophytes;			**Acute & Chronic RA PASSES at FOCUS Step 1 (algae) & FOCUS Step 2 (macrophytes)**
Lemna minor	$EC_{50(frond no.)}$	= 12 mg/L	
HMPA	$EC_{50(frond no.)}$	≥ 123 mg/L	
MON 52276	$EC_{50(frond no.)}$	= 21 mg/L	
Myriophyllum aquaticum	$EC_{50(fresh weight)}$	= 12.3 mg/L	
AMPA	$EC_{50(root length)}$	= 31.1 mg/L	
MON 52276	$EC_{50(fresh weight)}$	= 4.44 mg/L	

PECsw (worst case); FOCUS Step 1 = 491 µg/L
Step 2 N & S EU = 234.7 µg/L
Step 3 global max = 11.102 µg/L

Soil organisms incl. soil microbes

// Chronic – Technical material
// *56 day* repro (*E.fetida*). NOEC > 1000 mg/kg dw soil (equiv. 750 kg/ha)

// Soil macro-organisms (MON 52276)
// *H. aculeifer (repro)* 14 d NOEC = 473 mg/kg soil dw
// *F. candida (repro)* 28 d NOEC = 587 mg/kg soil dw

// Soil macro-organisms (AMPA)
// *H. aculeifer (reproduction)* 14 d NOEC = 320 mg/kg soil dw
// *F. candida (reproduction)* 28 d NOEC = 315 mg/kg soil dw

// Soil microbial - Nitrogen trans. (<25% diff. vs. control after 28 days)
// a.s. 6% difference after 28 days @ 33.1 mg/kg soil dw
// formulation 8% difference after 28 days @ 94 mg/kg soil dw
// AMPA 21% difference after 28 days @ 160 mg/kg soil dw

Expected RA outcome;

Based on max rate = 2.88 kg/ha;

Earthworm chronic

- PASS with TER >>5

Supported by low risk stated in EFSA (2015)

Soil macro-organisms

- PASS with TER >>5

Soil microbes

- 5 fold safety factor, based on worst case rate used to determine PECsoils.

PECsoil max = 3.84 mg/kg soil dw (based on 2.88 kg/ha rate)

Non-Target Terrestrial Plants (NTTPs)

// **Vegetative vigour**

ER_{50} = 28.4 g a.s./ha

// **Seedling emergence** (MON 52276)

ER_{50} > 2.88 kg a.s./ha

- // No soil herbicidal activity – supports historical position
- // Veg vig endpoints drive **NTTP assessment**

- // Based on ground directed applications at 2.9 kg/ha for weed control
  - // Off-target drift (1m) = 2.77%, equiv. = 79.8 g a.e./ha

Expected RA outcome

Anticipate PASS with limited refinement

Deterministic approach (10 species TER = 5)

- TER >5 (90% drift reducing technology + 1 m in-field buffer).

Probabilistic approach (SSD / HC_5) also possible

- achieves reduced level (%) DRT

New Studies since last Annex I renewal

New studies since last renewal

New studies

- **Broad leaf weed – residue decline** study in leafy crop – **IN DRAFT – see later slide**
- **Chironomous** 28 day developmental study – water spiked – **IN DRAFT**
- **Bee 22 day larval toxicity** study; on going **report expected Q3/Q4, 2019**.
- **Solitary bee** – acute contact toxicity - **complete**
- **Bumble bee** – acute oral + contact - **complete**
- **Honey bee** – 10 day laboratory feeding study - **complete**

Mammalian acute and chronic risk assessment – *key areas of the risk assessment;*

- Endpoint selection
- Other refinement Options

Endpoint selection - Acute Mammal Risk Assessment

- **EFSA (2015) acute mammal risk assessment endpoint;**

  >2000 mg/kg bw; lowest LD_{50}

- Unique position - 27 acute rodent toxicology oral gavage LD_{50} values available in current dataset.

- All LD_{50} effectively achieve the same outcome.
  - The LD_{50} values are greater than the highest dose selection in each study
    - AIR2 approach was to select the most frequently occurring LD_{50} (> 5000 mg/kg bw)

- Endpoint selection for AIR5 considers all available data;

Endpoint selection - Acute Mammal Risk Assessment

No	Species/ strai	Animal/dose level (mg/kg bw)	Vehicle	LD_{50} (mg/ kg bw)	Reference
1	Rat/ Sprague Dawley	5/sex/2000	Cotton seed oil	>2000	[redacted] 1995 Glyphosate Monograph.
2	Rat/ Spr. Dawley	1/sex/2000 5/sex/2000	Arachis oil	>2000	[redacted] 1994 Glyphosate Mono.
3	Rat, Wistar	5/sex/0 5/sex/5000	water	> 5000 (limit test)	[redacted] 1994 Glyphosate Mono.
4	Rat, Spr. Dawley	2/sex/250 2/sex/500 2/sex/1000 2/sex/3000 2/sex/5000 5/sex/5000	CMC	> 5000 (limit test)	[redacted], 1995 Glyphosate Mono.
5	Rat, Spr. Dawley	1/sex/2000 5/sex/2000	water	> 2000 (limit test)	[redacted], 1992 Glyphosate Mono.
6	Rat, Wistar	5/sex/2500 5/sex/5000 5/sex/7500	Peanut oil	> 7500 (estimated)	[redacted], 1991 Glyphosate mono.
7	Rat, CD	5/sex/0 5/sex/3000 5/sex/5000 5/sex/8000	1% CMC	> 8000	[redacted] 1990 Glyphosate Mono.
8	Rat, Spr. Dawley	5/sex/5000	0.5 % CMC	> 5000 (limit test)	[redacted] 1989 Glyphosate Mono.
9	Rat, Spr. Dawley	5/females/5000	water	> 5000 (limit test)	[redacted], 2009 Glyphosate renewal
10	Rat, Sprague Dawley	5/sex/5000	0.5 % CMC	> 5000 (limit test)	[redacted] 1995, Glyphosate Renewal
11	Mice, ICR	5/sex/5000	0.5 % CMC	> 5000 (limit test)	[redacted], 1995 Glyphosate renewal
12	Rat, Wistar	3 females/2000 (step 1) 3 females/2000 (step 2)	water	> 2000	[redacted], 2009 Glyphosate renewal
13	Rat, CD	3 females/2000 (step 1) 3 females/2000 (step 2)	0.8 % Hydroxy-propyl methylcellulose	> 2000 (limit test)	[redacted], 2009 Glyphosate renewal

GTF 2

Endpoint selection - Acute Mammal Risk Assessment

No	Species / strain	Animal/dose level (mg/kg bw)	Vehicle	LD_{50} (mg/ kg bw)	Reference
14	Rat, CD	3 females/2000 (step 1) 3 females/2000 (step 2)	0.8 % Hydroxy-propyl methyl cellulose	> 2000 (limit test)	[redacted], 2010a Glyphosate renewal
15	Rat, CD	3 females/2000 (step 1) 3 females/2000 (step 2)	0.8% Hydroxy-propyl methyl cellulose	> 2000 (limit test)	[redacted], 2010b Glyphosate renewal
16	Rat, Sprague-Dawley	3 females/5000	water	> 5000 (limit test)	[redacted], 2005 Glyphosate renewal
17	Rat, Wistar	3 females/2000 (step 1) 3 females/2000 (step 2)	water	> 2000 (limit test)	[redacted] 2008, Glyphosate renewal
18	Rat, HanRcc: WIST	2 x 3 ♀ x 3	PEG 300	> 2000 (limit test)	[redacted] 2007, Glyphosate renewal
19	Rat, Sprague Dawley	5/sex/5000	water	> 5000	[redacted], 1988 Glyphosate renewal
20	Rat, Wistar	5/sex/2500 5/sex/3500 5/sex/5000 5/sex/7000 5/sex/9900	water	> 5000	[redacted] 1979 Glyphosate renewal Suppl.
21	Rat	5/sex/5000	water	> 5000	[redacted] 1996 Glyphosate renewal
22	Rat	3 ♀ tpho	water	> 5000	[redacted], 2007 Glyphosate renewal
23	Rat	3 ♀ tpho	0.5 % CMC	>5000	[redacted], 2011 Glyphosate renewal
24	Rat	5 ♀ tpho	DMS	> 2000 (fixed dose)	[redacted], 2014 Glyphosate renewal
25	Mice, Swiss albino	5/sex/2500 5/sex/5000 5/sex/7500	Peanut oil	> 7500	[redacted], 1991 Glyphosate Mono.
26	Mice, Charles River	5/sex/2000	0.5 % CMC	> 2000 (limit test)	[redacted].,1994 Glyphosate Mono.
27	Mice, Bom: NM RI	5/sex/2000	water	> 2000 (limit test)	[redacted] 1991 Glyphosate Mono.

Endpoint selection - Acute Mammal Risk Assessment

- **Based on the 2000 mg/kg bw endpoint; Previous Annex I position;**
  - For 2.16 kg/ha rate & 90th%tile SV RUD value = 136.4; daily dietary dose (DDD) = 294.6,
    - TER 6.8 (<10). Therefore further consideration required as options to **refine acute mammal assessment limited**.
- **How..?**
  - Use most **frequently occurring LD_{50} value** (>5000 mg/kg bw)
  - Use **a derived endpoint** that considers all data – geomean endpoint **(3906 mg/kg bw)**
    - **Geometric mean acute LD_{50}** base on all 27 acute oral rodent studies
    - Acceptability of using geomean approach
      - Acute test designs considered substantially equivalent.
      - Clinical observations observed in acute oral studies;
        - **Transient** and **not sustained** for longer than 2-3 days.
        - All animals appeared **normal at end of 14 day observation period**.
    - **Acute risk assessment should be regulated based on lethality endpoint LD_{50}.**
      - In all cases the **'LD_0 was greater than the highest dose.'**

Endpoint selection - Acute Mammal Risk Assessment

Proposals by GTF2

- To demonstrate substantial equivalence in acute oral rodent study designs.
- Use an alternate acute mammalian endpoint in the assessment

Question to AGG

- Do the AGG agree with the proposed approach to use an alternate endpoint in the acute risk assessment ?

- Both are considered conservative considering all LD_0 were > highest dose tested
  - i.e. **no mortality observed across all studies**.

Endpoint selection – Chronic Mammal Risk Assessment

- **AIR2; Chronic mammalian endpoint for use in risk assessment = 50 mg/kg bw/day**
  - rabbit developmental toxicology study

- Endpoint selection is considered conservative, why..?
  - Size of available dataset – Multiple rabbit dev tox studies available
    - only the lowest endpoint is considered
  - Higher NOAELs in dataset that are below the lowest relevant LOAEL.
  - Chronic oral gavage does not reflect dietary exposure in field

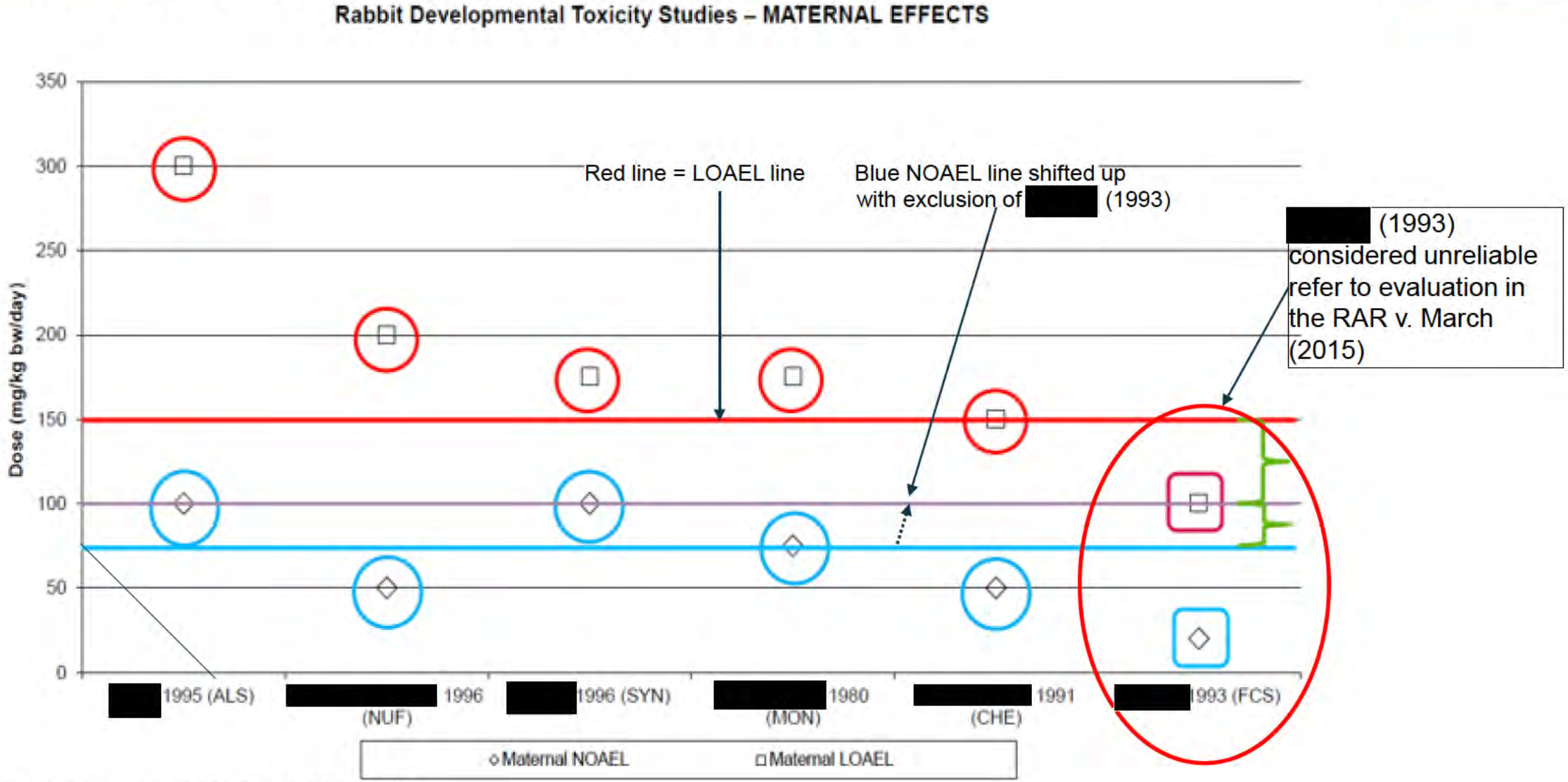
- **Relevance of rabbit dev tox endpoint**
  - Clinical signs considered to be due **to G.I. tract irritation**
  - Soft stools / liquid faeces – concern for coprophagous animals being nutritionally compromised
    - **malnutrition**

Endpoint selection – Chronic Mammal Risk Assessment

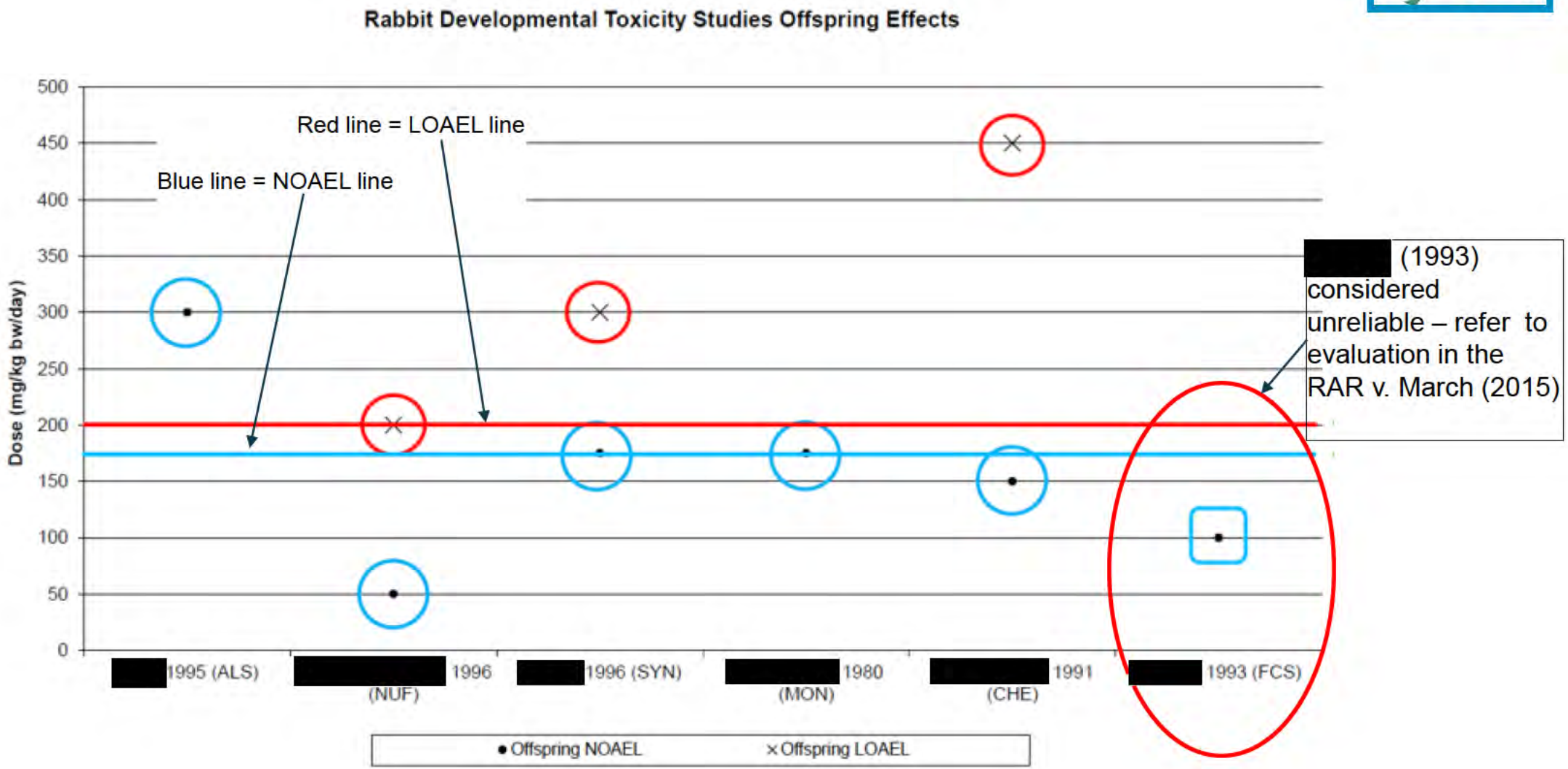
- **Multiple 'reliable' rabbit developmental toxicology studies available (RAR, rev. March 2015);**
  - [redacted] **(1996)**: Acceptable; 50, 200, 400 mg/kg/d - Dosed on gestation days (GD) days 7-19
  - [redacted] **(1996)**: Acceptable; 100, 175, 300 mg/kg/d - Dosed GD days 8-20
  - [redacted] **(1995)**: Acceptable; 10, 100, 300 mg/kg/d- Dosed GD days 6-18
  - [redacted] **(1991):** Acceptable; 50, 150, 450 mg/kg/d - Dosed GD days 7-19
  - [redacted] **(1980):** Supplementary (high maternal mortality & ↓to 6 litters @ high dose); 75, 175, 350 mg/kg/d
    - Dosed GD days 6-27
  - [redacted] (1993): Supplementary; 20, 100, 500 mg/kg/d
    - Dosed GD days 6-18 (*- unreliable study as evaluated in EU RAR rev. March, 2015*)
  - Assessment of **maternal** and **developmental / offspring** effects

Endpoint selection – Chronic Mammal Risk Assessment

- EFSA guidance document (2009) section 2.4.3, approach combine study results as if generated in one study.
  - Achieved study endpoints presented graphical allows for clear interpretation



Endpoint selection – Chronic Mammal Risk Assessment



Endpoint selection – Chronic Mammal Risk Assessment

- **Maternal** Effects;
  - Lowest **LOAEL = 150 mg/kg bw/day**

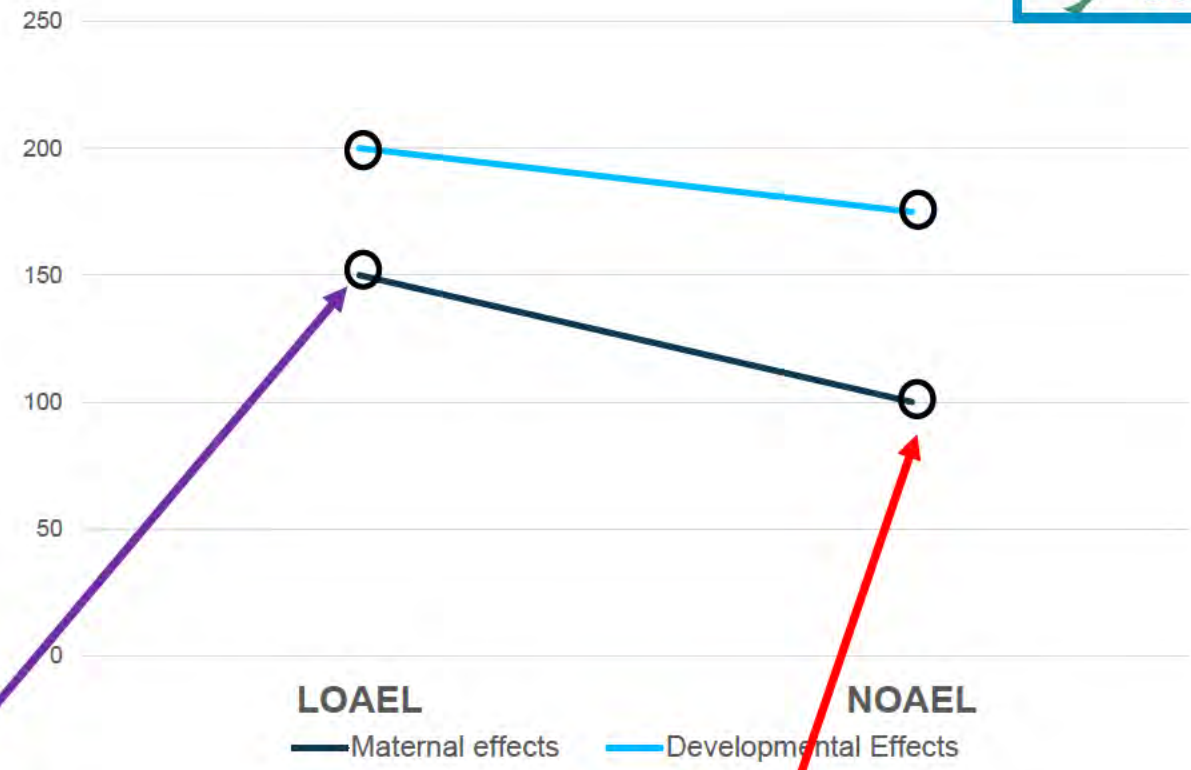
    [redacted] (1991)
  - Highest **NOAEL = 100 mg/kg bw/day**

    [redacted] (1995) & [redacted] (1996)

- **Developmental (offspring)** Effects;
  - Lowest LOAEL = **200 mg/kg bw/day**

    [redacted] (1996)
  - Highest NOAEL = **175 mg/kg bw/day**

    [redacted] (1996) & [redacted] (1980)



Highest NOAEL below LOWEST relevant LOAEL

= 100 mg/kg bw/day

<u>However – what about the nature / severity of effects at maternal LOAEL.. ?</u>

- Are these relevant at the population level..?

Endpoint selection – Chronic Mammal Risk Assessment

- Firstly – consider protection goals in EFSA (2009) relevant at the **population level**..?
  - **EFSA (2009)**

    '..surrogate protection goal of making any <u>mortality</u> or <u>reproductive</u> effect unlikely.'

    - considered at 1st Tier of chronic mammal risk assessment.
- What effects are relevant to wild mammal assessment at <u>population level</u>;

Both…

- **<u>Mortality</u>**;
  - ***Individual maternal or offspring mortality***

    - individual mortality; **<u>unacceptable</u>**
- **<u>Reproductive effect</u>;**
  - ***Implantation losses***
  - ***Reduced offspring number***
  - ***Non-viable offspring***

    - generational impact effecting both abundance & diversity; **<u>unacceptable</u>**

Endpoint selection – Chronic Mammal Risk Assessment

- **Some studies the <u>Maternal LOAEL</u> and <u>Developmental NOAEL</u> occur at the same dose..**
  - In [redacted] (1996), Maternal **LOAEL** & Developmental **NOAEL** = 175 mg/kg bw/day
  - In [redacted] (1991), Maternal **LOAEL** & Developmental **NOAEL** = 150 mg/kg bw/day

- **No maternal mortality nor developmental (offspring) effects** at the <u>maternal **LOAEL**</u>
  - Observed maternal clinical effects at LOAELs of <u>150 & 175 mg/kg bw/day</u> were;
    - appetite loss
    - reduced body weight gain
    - soft stools / liquid faeces
    - <u>No implantation losses at these dose levels..!</u>
      - Lowest rate with implantation losses = 200 mg/kg bw/day '$LOAEL_{developmental}$ ([redacted], 1996)'
  - Based on '**actual protection goal**' (EFSA (2009) applicable at refinement step of assessment;
    - '*..no visible mortality or long-term repercussions on abundance and diversity.*'
  - Endpoint **NOAEL** of 150 mg/kg bw day - considered protective of **<u>both</u> maternal and developmental effects** at the population level for wild mammals, meeting the EFSA (2015) protection goal.

Endpoint selection – Chronic Mammal Risk Assessment

Proposals by GTF2

Use **alternate endpoint from rabbit developmental toxicology endpoints** that is considered relevant at wild mammal population level.

e.g.

Based on the 150 mg/kg bw/day endpoint;

2.16 kg/ha, RUD = 72.3, fTWA of 0.19.

Daily dietary dose = 29.7; **TER = 5.05** (TER trigger >5)

- Proposal aims to support **all uses.**
- **Additional information to be presented in tox meeting that could enable alternate species endpoint use in chronic mammal risk assessment.**

Question to AGG

- What is AGG opinion to the above approach ?
  - *If the alternate endpoint proposal is not accepted then additional higher tier studies may be required e.g. see enclosure study proposal in later slides*

Refinement options for use in Bird & Mammal Risk Assessments

Refinement options applicable to Bird and Mammal Risk Assessments – refinement options

- **AIR2 submission included;**
  - **Grass residue decline data** - DT_{50} for grasses / 2.8 days modified fTWA = 0.19 for chronic assessment
  - **Ground arthropod residue decline & Broadleaf plant residue decline data**.
    - Suggested DT_{50} values for glyphosate decline on ground arthropods & broadleaf plants of 3-6 days.
    - Study acceptance at zRMS and national level mixed - concerns few locations and sample sizes.
  - **Access to broadleaf plant residue decline study conducted in pea (surrogate broadleaf weed) - multiple countries (N & S EU residue zone); Rate =** 2.88 kg/ha
  - Prelim results indicate
    - Plants were highly susceptible
    - Succumb within 1-4 days
      - Limited value of data in RA
      - Ongoing evaluation within GTF2.

Results including 21 day TWA concentrations and f_{twa} values

Dataset	Model	Half-life [days]	Fitted initial residue [mg/kg]	21 day TWA [mg/kg]	f_{twa} [-]
Trial 1	SFO	4.3	148	42.3	0.286
Trial 2	SFO	7.19e+10	87.3	87.3	1
Trial 3	HS	-	118	35.7	0.304
Trial 4	SFO	1.7e+10	116	116	1
Trial 5	FOMC	-	100	54.1	0.54
Mean			**114**	**67.1**	**0.626**

Higher Tier Common vole semi-field effects study

- **Higher tier refined approaches, may be required to support certain uses e.g.;**
  - Population modelling
    - current e-vole modelling - acceptability considered limited
    - Submission of current modelling in discussion with GTF2 membership
- Field studies with wild caught mammals
  - Semi-field enclosure study to support orchard uses
    - **<u>At least required where alternate endpoint scenarios are not considered appropriate.</u>**

- Field study refinement - for small herbivorous mammals; options are limited.. 100% grass in diet / high default RUD
  - **Possible to conduct a study with vole in** Spring 2020 – this would mean report submission after dossier submission in June 2020
    - For orchard uses a banded or strip application of product to control weeds between trees would be simulated – applied within tree rows around tree trunks - reduced total applied area (50%) in RA EFSA (2015).

Higher Tier Common vole semi-field effects study

- Possible test design to support uses in Top fruit / orchard / vineyards
  - A semi-field enclosure study using a banded / strip application made using shielded sprayer - simulating ‘orchard use’.
  - Possible exposure rates; **1.44 and 2.88 kg/ha** + control
    - **Four** replicate enclosures per treatment and control group.
  - Monitoring of wild caught and released vole populations in enclosure using
    - Mark-recapture trapping techniques and wildlife cameras to monitor behaviours / wilting times.
  - End of study - animals will be released at parental capture locations.

Higher Tier Common vole semi-field effects study design

- Farmland (arable) site
- 3 mm stainless steel sheets buried 1 m into ground and 50 cm above ground to avoid escapes / migration of voles by burrowing or jumping. Farmland site
- 12 enclosures; 15m x 15m (225 m^2 per enclosure)
- Entire enclosure covered with netting to avoid bird and mammal predators.
- Ground cover will be mixed grasses to simulate orchard under-storey
- Populations monitored using Ugglan multi-capture live traps for 'mark / recapture' techniques in each enclosure
- Starting 'population' at least 4 individually marked voles

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Higher Tier Common vole semi-field effects study

Proposals by GTF2

- A semi-field enclosure study **could** be conducted in 2020 to support use in orchard type crops
  - ***Especially where alternate endpoint strategies are not considered appropriate.***
- A report from or such a study would be submitted after June 2020 dossier submission due to seasonality of voles
- All relevant / reliable glyphosate residue decline studies will be submitted in June 2020

Question to AGG

- Will the AGG accept a 50% reduction in rate applied for use in the risk assessments for perennial row crops considering banded application ?
- Do the AGG agree to review the available residue decline studies – some whch have been previously reviewed at EU level ?
- Where broadleaf data set no considered useful for EU RA, what options are available to RA.. e.g. Can grass decline data be applied across all plant matter in diet ?
- Do the AGG agree that an enclosure study as described would be considered an acceptable refined approach to support orchard uses?

Aquatic Risk Assessment

- chronic Endpoint Selection

Aquatic Risk Assessment Endpoint selection / positioning

- Chronic fish endpoint 1000 µg/L (*Brachydanio rerio* sac fry assay (OECD 212).
  - Study design used as a pre-study for ELS tests – not considered robust for risk assessment
    - poor temperature maintenance / temperature shock / handling stress
    - control validity criteria not satisfied - swim-up not possible to confirm as free swimming larvae were exposed
    - no feeding during 168 hour duration - starvation cannot be ruled out as having influenced outcome of study.
    - test media not analysed.

- **Species selection** – chronic fish test - not performed on most acutely sensitive fish species;
  - Zebrafish acute 96hr LC_{50} = 123 mg/L; Rainbow trout acute 96hr LC_{50} = 38 mg/L

- **Glyphosate dataset** contains more relevant chronic exposure studies;
    - Full fish life cycle study / fathead minnow / NOEC 25.7 mg/L
    - Fish early life stage test / rainbow trout / NOEC 9.6 mg/L.
- If AGG agree that study not fit for use in risk assessment – would affect the CLP classification

Aquatic Fish Chronic endpoint selection

Proposals by GTF2

- The rainbow trout fish early life stage endpoint (9.6 mg/L) to be used in the fish chronic risk assessment.
- Argumentation to be presented in dossier to support exclusion of the *Brachydanio rerio* endpoint use in risk assessment.

Question to AGG

- Do the AGG agree with the proposed approach..?

Pollinators

Pollinators: Available non-Apis & Apis sp. studies & risk assessment

- **AIR2 submission;**
- Honeybee acute oral/contact with technical material and representative formulation
  - supported by Honeybee colony feeding study – also evaluated during AIR2.
  - EFSA (2015) concluded *'A low risk was concluded on first tier risk assessments for bees, non-target arthropods….'*
  - Annex I risk assessment used **EPPO** approach / HQs Annex VI trigger = 50.
- Additional studies
  - Chronic honey bee 10 day lab & non – *Apis sp.* acute studies conducted anticipating release of the EFSA Bee Guidance document, including 22 day chronic honey bee larval study (in-draft).

- **2019 has seen changes to;**
  - Regulation (EC) No. 1007/2009, Annex VI Uniform Principles for the Protection of Bees / trigger values
  - European Commission roadmap for the implementation of the EFSA bee guidance document,
    - = delayed implementation of guidance document.
  - Overall - risk assessment now to be based on results of acute studies / new triggers + weight of evidence

Pollinators: Chronic effects of glyphosate on Honey bee larvae

Proposals by GTF2

- To complete and submit a Honey Bee Larval Toxicity Test, Repeated Exposure study.
- To submit non-Apis acute studies and the 10 day honey bee adult feeding study, as part of weight of evidence to support existing risk assessment.

Question to AGG

- Please could the AGG provide guidance on how they would like the chronic bee data to be presented in the risk assessment ?
  - E.g. as a weight of evidence – similar to current Article 43 submissions or differently ?

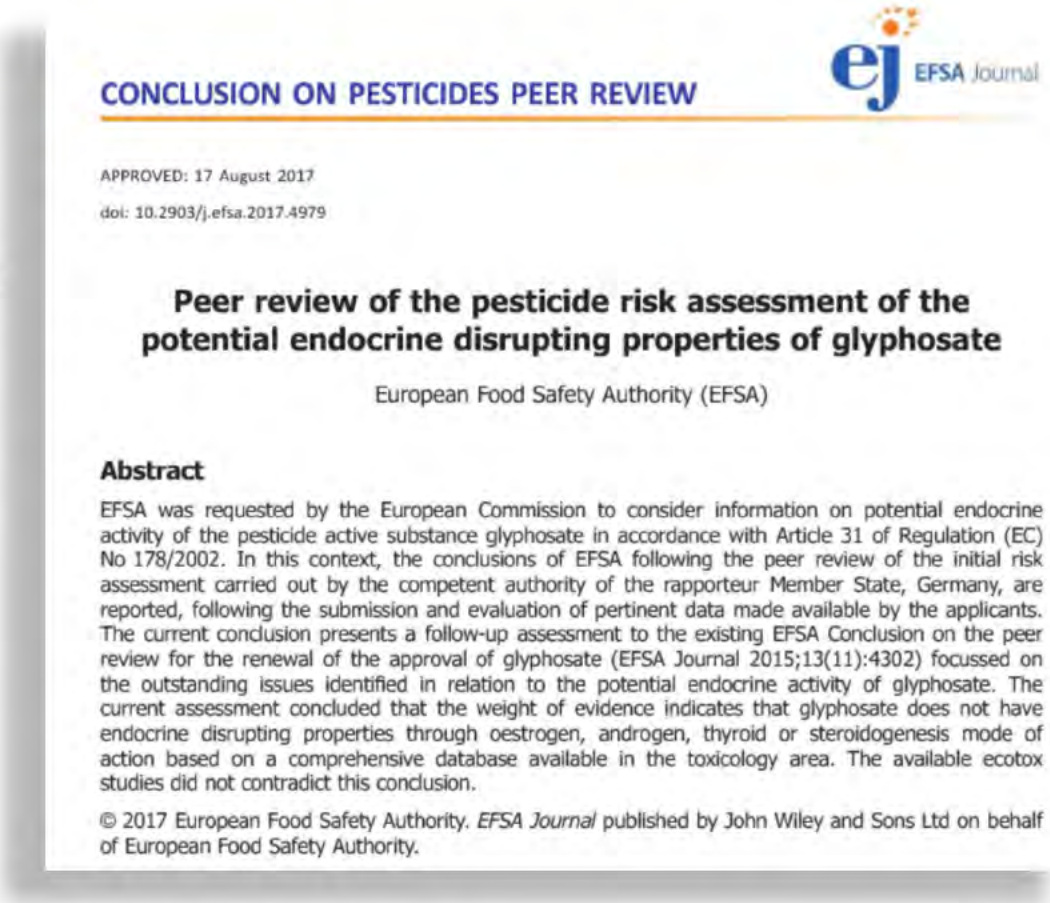
Endocrine Disruption

Endocrine Disruption & Glyphosate

GTF 2

- **EFSA conclusion on endocrine disruption;**

- *'All the experts agreed that the weight of evidence indicates that **glyphosate** does not have **EATS-mediated endocrine disrupting** properties and that the data gap identified in the previous **EFSA conclusion** (**EFSA**, 2015) has been adequately addressed.'*



CONCLUSION ON PESTICIDES PEER REVIEW

EFSA Journal

APPROVED: 17 August 2017

doi: 10.2903/j.efsa.2017.4979

Peer review of the pesticide risk assessment of the potential endocrine disrupting properties of glyphosate

European Food Safety Authority (EFSA)

Abstract

EFSA was requested by the European Commission to consider information on potential endocrine activity of the pesticide active substance glyphosate in accordance with Article 31 of Regulation (EC) No 178/2002. In this context, the conclusions of EFSA following the peer review of the initial risk assessment carried out by the competent authority of the rapporteur Member State, Germany, are reported, following the submission and evaluation of pertinent data made available by the applicants. The current conclusion presents a follow-up assessment to the existing EFSA Conclusion on the peer review for the renewal of the approval of glyphosate (EFSA Journal 2015;13(11):4302) focussed on the outstanding issues identified in relation to the potential endocrine activity of glyphosate. The current assessment concluded that the weight of evidence indicates that glyphosate does not have endocrine disrupting properties through oestrogen, androgen, thyroid or steroidogenesis mode of action based on a comprehensive database available in the toxicology area. The available ecotox studies did not contradict this conclusion.

Endocrine Disruption & Glyphosate

- **Evaluation for AIR 2 included;**
  - ED specific literature search – publication period covered: January 2014 to October 2016[a]
    - Weight of evidence analysis of EDSP Tier 1 screening assays[b]
      - Including the US EPA memorandum outlining evaluation and conclusion of EDSP Tier 1 screening assays[c]
    - EFSA peer review on the potential endocrine activity of glyphosate[d]:
- Based on available ecotoxicology studies - glyphosate has no androgenic, estrogenic, steroidogenic or thyroidal effects

For the AIR5 Evaluation to fulfil EU ED requirements[e]:

- Update of ED specific literature search - publication period covered: November 2016 to July 2019

- Evaluation of existing study data according to EU requirements;
  - 4 x avian reproduction studies (two are according to OECD 206).
  - 1 x fish full life cycle (EPA methodology, 1971).
  - 1 x fish early life stage (OECD 210).
  - 1 x US EDSP Tier 1 - Fish short-term reproduction assay (OECD 229).
  - 1 x US EDSP Tier 1 - Amphibian metamorphosis assay (OECD 231).

[a] Literature search on Glyphosate. Glyphosate Task Force, 2016.
[b] Weight of evidence analysis for Glyphosate: An evaluation of results from the EDSP Tier 1 screening assays. Glyphosate Task Force, 2016.
[c] EDSP: weight of evidence analysis of potential interaction with the estrogen, androgen or thyroid pathways - Glyphosate. US EPA, 2015.
[d] Peer review of the pesticide risk assessment on the potential endocrine activity of glyphosate. EFSA Journal 2017;15(9):4979.
[e] Guidance for the identification of endocrine disruptors in the context of Regulations (EU) No 528/2012 and (EC) No 1107/2009 EFSA Journal 2018;16(6):5311)

Endocrine Disruption & Glyphosate

- **US EDSP (Endocrine Disruption Screening Programme) review;**
  - The existing US ED assessment will be adapted to meet the mandatory EU ED assessment (ED GD, 2018)
  - **That will include;**
    - Comprehensive ED targeted literature search / Filling in Table Appendix E (ED GD template)
    - Integration of Lines of Evidence / Reporting in Appendix I
    - Conclusions
  - **Preliminary conclusions;**
    - **No ED concern from available studies**
      - No further testing deemed required due to extensive dataset and results from available studies indicating no further ED concern

Endocrine Disruption

<u>Overall Conclusion</u>: glyphosate is not considered to be an endocrine disruptor

(BfR, 2017 – Glyphosate Addendum 2 to RAR)

Study Type	Assays (OECD Level)	Results
In vitro	ER Binding (2) ER Transcriptional Activation (2) AR Binding (2) Steroidogenesis H295R (2) Aromatase (2)	The weight of evidence indicates that glyphosate does not have potential to interact with the EATS-mediated endocrine pathways*
In vivo	Uterotrophic (3) Hershberger (3) Pubertal Male (4) Pubertal Female (4) Amphibian Metamorphosis (ECO) Fish Short-term Reproduction(ECO)	

* Revisit molecular structure and pharmaco-kinetics

Endocrine Disruption Conclusion

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Keywords: glyphosate, peer review, potential endocrine activity, risk assessment, pesticide, herbicide

Requestor: European Commission

Question number: EFSA-Q-2016-00663

Correspondence: pesticides.peerreview@efsa.europa.eu

*The weight of evidence indicates that **glyphosate is not an ED.***
EFSA – September 2017

Submission will follow EFSA guidance on endocrine disruption assessment in Appendix E.

Endocrine Disruption – preparation of Appendix E

OECD Conceptual Framework			
Level 1	Existing data and new-nontesting information		
Level 2	In vitro assays on selected endocrine mechanisms	5 EDSP assays	Estrogen Receptor(ER) Binding Assay
			ERα Transcriptional Activation assay
			Androgen Receptor (AR) binding assay
			Steroidogenesis
			Aromatase Inhibition Assay
Level 3	In vivo assays on selected endocrine mechanisms	2 EDSP	Hershberger Assay
			Uterotropic assay
Level 4	In vivo assays on adverse selected selected endocrine mechanisms	2 EDSP	Pubertal developmental and Thyroid function in male rats
			Pubertal developmental and Thyroid function in female rats
		In vivo toxicity studies	70 studies with rats, mice, dogs and rabbits
Level 5	In vivo assay covering life cycle changes		6 Two-generational reproductive toxicity

Question to AGG

Given the significant quantity of higher tier data available, does the AGG consider Level 1 data unnecessary?

Biodiversity

Biodiversity

Biodiversity – currently not a specific data requirement under Regulation (EC) No. 1107/2009

- Biodiversity in the Regulation (1107/2009) in Chapter II, Section 1, subsection 1, Article 4: Approval criteria for active substances, paragraph 3, where it states for Active substances…
  - *…' shall have no unacceptable effects on the environment, having particular regard to the following considerations where the scientific methods accepted by the Authority to assess such effects are available: …(iii) its impact on biodiversity and the ecosystem.'*

- No EU wide guidance yet adopted for assessing the impact of plant protection products on biodiversity through trophic interactions in the environment at a landscape level.
  - 'Biodiversity' , 'direct effects' and 'indirect effects' are amongst the search terms used in lit review.

Question to AGG  Can the AGG advise the GTF as to how biodiversity should be addressed in the dossier ?

Biodiversity

For example;

It is anticipated that the risk assessment in the dossier will support low risk conclusions based on uniform principles – that align with Regulation (EC) No. 1107/2009 protection goals.

- Considering;
  - Direct effects – based on ecotoxicity studies and exposure estimates.
  - Indirect effects – based on secondary poisoning (birds / mammals) via drinking water / dietary items / bioaccumulation along the food chain.
  - Additionally, glyphosate = good environmental profile due to favourable phys/chem properties;
    - High Koc limiting mobility in soils, with rapid dissipation to sediments if It reaches water
    - FOCUS Modelling predicts low surface water concentrations (Aquatic RA passes at FOCUS Step 2/3)
    - **AIR5 propose GAP use rates = substantially lower application rates than AIR2**

Question to AGG 	***Would a qualitative review of the potential impacts of glyphosate on biodiversity through trophic interactions, be sufficient to inform on the risk assessment / AGG evaluation ?***

Thank you!

Biodiversity

- A large margin of safety anticipated for soil organism exposure based on AIR2 / Article 43 evaluations
  - Earthworms / predatory soil mites / soil detritivores & soil microbes – low risk

- Glyphosate transfer through food chain = negligible (Log Kow<1, BCF factor of 1.1 (trigger 1000))

- Rapid excretion from bodies of organisms expected via urine and faeces with limited metabolism.

- Foliar residues of glyphosate on plants – rapid decline expected (<5 days) reduces dietary exposure of non-target animals / loss of palatability, expect exposure time in field = short.

- Non-target arthropod populations on developing crop leaves will be mostly unaffected – maintaining in-field populations of beneficial arthropods due to ground directed applications.

Biodiversity – Overall statement

- Glyphosate application will lead to indirect effects on non-target organisms as product applied to control unwanted plants in modified / managed landscapes.
  - No suitable risk assessment available to address both landscape or agronomic management system' questions beyond the ecotoxicity profile.
    - Risk assessment = direct ecotoxicological effects – which includes secondary poisoning via trophic interactions.
- Habitat provision; 'in-field and off-field' – depends on species specific strategies.
- Species and ecosystems of conservation concern in Europe, depend on historical agricultural practices and extensive management.
  - Abandonment such systems considered as much a threat to biodiversity as intensification.
- Out of production schemes
  - field margins / hedgerows are effective at enhancing species richness
    - Implementation requires decision at landscape level.
- Boosting yields on farmland / sparing natural habitats would reduce impact on wildlife populations.
  - e.g. use of precision farming techniques

Biodiversity – other points that will feature in GTF position on Biodiversity

- Biodiversity should be managed at the **landscape / policy level**, requiring alignment of agriculture and nature conservation policies which are considered outside of the Regulatory / PPP framework.
- A review of the effects of glyphosate on plant & animal diversity in managed systems reported that **changes in species richness & diversity of plants, birds & small mammals** were within the **range of natural fluctuations**.
- **Glyphosate** enables adoption of **no- or low-till tillage practices**, which promote soil biodiversity and water quality and reduced-tillage practices are an important part of sustainable agriculture.
- **Glyphosate** is a **core element** of **all Integrated Weed Management (IWM) programs in Europe**.
- **Glyphosate** is used to **restore habitats and control invasive plant species**. Its an important tool to protect EU biodiversity under the **new EU Alien Invasive Species regulation** due to broad mode-of-action being ideally suited to control many invasive plant species + a favorable environmental safety profile.

Biodiversity

- Glyphosate use is a critical part of modern sustainable agriculture providing benefits and minimal risk to the environment.
  - Primary benefits include;
    - Enabling tillage practices that reduce soil erosion, improving soil quality and biodiversity.
    - Providing high efficacy against a broad spectrum of agriculturally and non-agriculturally important weeds as well as a tool to control alien invasive species.
    - Use of glyphosate enables conservation measures, which have a higher potential to sustain regional above-ground carbon stocks compared to other strategies (Williams *et al.*, 2018).
  - The current risk assessment process performed according to the Uniform Principles of the Regulation (EC) No. 1107/2009 are therefore considered to be acceptable. This considers direct exposure effects of glyphosate. The anticipated results of the risk assessment, are considered protective of non-target organisms occurring both in-field and in off target areas.