

ADOPTED: 7 October 2019

doi: 10.2903/j.efsa.2019.5888

Safety for the environment of Monimax[®] (monensin sodium and nicarbazin) for chickens for fattening, chickens reared for laying and for turkeys for fattening

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Abstract

Following a request from the European Commission, the Panel on Additives and Products or substances used in Animal Feed (FEEDAP Panel) was asked to deliver a scientific opinion on the safety and efficacy of the coccidiostat Monimax[®] (monensin sodium and nicarbazin) when used in feed for turkeys or chickens for fattening. In previous assessments (2017, 2018), the FEEDAP Panel could not conclude on the safety of Monimax[®] for the environment due to concerns on monensin sodium and 4,4'-dinitrocarbanilide (DNC, one of the moieties of nicarbazin). The applicant provided additional information that has been assessed. The use of monensin sodium from Monimax[®] in complete feed for chickens for fattening, chickens reared for laying and turkeys for fattening poses no risk for the aquatic compartment, the terrestrial compartment or for sediment. The bioaccumulation potential of monensin in the environment is low. No concerns would arise for the 4,6-dimethylpyrimidin-2-ol (HDP) moiety of nicarbazin excreted from chickens for fattening, chickens reared for laying and turkeys fed Monimax[®]. The use of DNC moiety of nicarbazin from Monimax[®] in complete feed for chickens for fattening, chickens reared for laying and turkeys for fattening poses no risk for the aquatic compartment, the terrestrial compartments or for sediment. The bioaccumulation potential of DNC in the environment is low and the risk for secondary poisoning is not likely to occur

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Keywords: coccidiostat, Monimax, monensin sodium, nicarbazin, safety, environment, chickens for fattening, turkeys for fattening

Requestor: European Commission

Question number: EFSA-Q-2018-00925 and EFSA-Q-2019-00253

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Acknowledgements: The Panel wishes to thank the following for the support provided to this scientific output: Jaume Galobart.

Suggested citation: EFSA FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed), Bampidis V, Bastos ML, Christensen H, Dusemund B, Kouba M, Kos Durjava M, López-Alonso M, López Puente S, Marcon F, Mayo B, Pechová A, Petkova M, Ramos F, Sanz Y, Villa RE, Woutersen R, Finizio A, Focks A, Teodorovic I, Tarrés-Call J, Vettori MV and Azimonti G, 2019. Scientific Opinion on the safety for the environment of Monimax® (monensin sodium and nicarbazin) for chickens for fattening, chickens reared for laying and for turkeys for fattening. *EFSA Journal* 2019;17(11):5888, 12 pp. <https://doi.org/10.2903/j.efsa.2019.5888>

ISSN: 1831-4732

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The EFSA Journal is a publication of the European Food Safety Authority, an agency of the European Union.



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1. Introduction

1.1. Background and Terms of Reference as provided by the requestor

Regulation (EC) No 1831/2003 establishes rules governing the Community authorisation of additives for animal nutrition and, in particular, Article 9 defines the terms of the authorisation by the Commission.

The applicant, Huvepharma NV, is seeking a Community authorisation of Monensin sodium and Nicarbazin as feed additive to be used as a Coccidiostat and histomonostat for chickens for fattening, chickens reared for laying and turkeys for fattening (Table 1).

Table 1: Description of the substances

Category of additive	Coccidiostat and histomonostats
Functional group of additive	
Description	Monensin sodium and Nicarbazin
Target animal category	Chickens for fattening, chickens reared for laying and turkeys for fattening
Applicant	Huvepharma NV
Type of request	New opinion

On 29 November 2017, the Panel on Additives and Products or Substances used in Animal Feed of the European Food Safety Authority ('Authority'), in its opinion on the safety and efficacy of the product, could not conclude on the safety on environment of monensin sodium¹ and nicarbazin as a feed additive for turkeys for fattening due to (i) uncertainties linked to the very high persistence of the compound 4,4'-dinitrocarbanilide (DNC) refined predicted environmental concentrations (PECs), (ii) DNC might accumulate in the sediment compartment and (iii) DNC can potentially bioaccumulate and may cause secondary poisoning.

On 14 November 2018, the Panel on Additives and Products or Substances used in Animal Feed of the Authority, in its opinion on the safety of the product, could not conclude on the safety of monensin sodium and nicarbazin in chickens for fattening and chickens reared for laying, under the conditions of use proposed by the applicant.

The Commission gave the possibility to the applicant to submit complementary information in order to complete those assessments and to allow a revision of Authority's opinion. The new data on turkeys for fattening were received on 26 October 2018, and the new data for chickens for fattening were received on 18 October 2018.

In view of the above, the Commission asks the Authority to deliver a new opinion on safety for environment of monensin sodium and nicarbazin as feed additive for chickens for fattening, chickens reared for laying and turkeys for fattening based on the additional data submitted by the applicant.

1.2. Additional information

The additive Monimax® contains as active substances monensin sodium and nicarbazin. Monensin sodium is a polyether ionophore produced by fermentation from a culture of *Streptomyces* spp. Nicarbazin, produced by chemical synthesis, is an equimolar complex of 1,3-bis(4-nitrophenyl)urea, also known as *N,N'*-bi(4-nitrophenyl) urea or 4,4'-dinitrocarbanilide (DNC) and 4,6-dimethylpyrimidin-2-ol (HDP).

Currently, the feed additive Monimax®, containing monensin sodium and nicarbazin as active substances, is not authorised in the European Union.

The FEEDAP Panel assessed the safety and efficacy of Monimax® for chickens for fattening, chickens reared for laying and turkeys for fattening in 2017 and 2018 (EFSA FEEDAP Panel, 2017, 2018) and could not conclude on the safety of the additive for the environment due to concerns for monensin sodium and DNC.

The applicant has provided additional information to demonstrate the safety of the additive for the environment.

¹ For monensin sodium a risk could not be excluded for the terrestrial compartment based on the results of an ecotoxicity test on plants (EFSA FEEDAP Panel, 2017, 2018).

2. Data and methodologies

2.1. Data

The present assessment is based on data submitted by the applicant in the form of additional information² in support of the authorisation request for the use of Monimax® (monensin sodium and nicarbazin) as a feed additive for chickens for fattening, chickens reared for laying and turkeys for fattening, following previous applications on the same product.³ The additional information received for chickens for fattening and chickens reared for laying is the same as the one received for turkeys for fattening.

2.2. Methodologies

The approach followed by the FEEDAP Panel to assess the safety for the environment of Monimax® (monensin sodium and nicarbazin) for chickens for fattening, chickens reared for laying and turkeys for fattening is in line with the principles laid down in Regulation (EC) No 429/2008⁴ and the relevant guidance documents: Guidance for assessing the safety of feed additives for the environment (EFSA, 2008).

3. Assessment

The product under application is Monimax®, containing 76–84 g monensin sodium and 76–84 g nicarbazin/kg product as active substances. It is intended to be used in feed for chickens for fattening, chickens reared for laying up to 16 weeks of age and turkeys for fattening up to 16 weeks of age as a coccidiostat at the recommended inclusion level of 40 + 40 to 50 + 50 mg monensin + nicarbazin/kg complete feed. The applicant proposes a withdrawal period of 1 day.

In 2017, the FEEDAP Panel issued an opinion on the safety and efficacy of Monimax® for turkeys for fattening (EFSA FEEDAP Panel, 2017) and, 1 year later, another opinion on the safety and efficacy of Monimax® for chickens for fattening and for chickens reared for laying EFSA FEEDAP Panel (2018). In its opinion on turkeys for fattening, the FEEDAP Panel concluded as follows:

The use of monensin sodium from Monimax® in complete feed for turkeys does not pose a risk for the aquatic compartment and sediment, while a risk cannot be excluded for the terrestrial compartment based on the results of an ecotoxicity test on plants. The bioaccumulation potential of monensin in the environment is low.

A final conclusion on the risk resulting from the use of nicarbazin from Monimax® in turkeys cannot be made for the following reasons: (i) DNC refined PECs showed uncertainties linked to the very high persistence of the compound, (ii) DNC might accumulate in the sediment compartment and (iii) DNC can potentially bioaccumulate and may cause secondary poisoning. The PEC/PNEC ratios indicate a risk for daphnids but no adverse effects were seen at the concentration tested. This adds further uncertainty to the risk assessment of DNC in the aquatic compartment. No concerns would arise for the HDP moiety of nicarbazin excreted from turkeys fed Monimax®.

The potential of DNC to accumulate in soil over the years should be investigated by monitoring in a field study.

In summary, based on the available data, the FEEDAP Panel cannot conclude on the safety of Monimax® for the environment.

The same conclusions were reiterated in its opinion on the safety and efficacy of Monimax® for chickens for fattening and chickens reared for laying (EFSA FEEDAP Panel, 2018).

The applicant has provided new data that are the object of the current assessment. In particular, a new ecotoxicity test on terrestrial plants with monensin sodium⁵ and data to address the uncertainties linked to the very high persistence of DNC for the environment.

² FEED dossiers references: FAD-2018-0079 and FAD-2018-0081.

³ FEED dossiers references: FAD-2012-0032 and FAD-2012-0027.

⁴ Commission Regulation (EC) No 429/2008 of 25 April 2008 on detailed rules for the implementation of Regulation (EC) No 1831/2003 of the European Parliament and of the Council as regards the preparation and the presentation of applications and the assessment and the authorisation of feed additives. OJ L 133, 22.5.2008, p. 1.

⁵ Technical dossier FAD-2018-0079/Supplementary information May 2019/References/Reference 1 to 4.

3.1. Safety for the environment

3.1.1. Monensin sodium

The characterisation of the risk for the terrestrial compartment for monensin sodium is updated in the current assessment based on the results of the new good laboratory practice (GLP)-compliant ecotoxicity study in plants performed according to the OECD guideline 208.⁵

In that new study, a natural sandy loam soil was treated with monensin (as monensin sodium) at seven concentrations and seeds from six species were sown (monocotyledon species *Hordeum vulgare* and *Allium cepa*, and dicotyledon species *Phaseolus vulgaris*, *Raphanus sativus*, *Cucumis sativa* and *Solanum lycopersicum*). The endpoints determined were the effects on emergence/survival, phytotoxicity, shoot length, and fresh weight and dry weight biomass. Dry biomass was the most sensitive endpoint for all species with *A. cepa* showing the highest sensitivity ($EC_{50} = 4.99$ mg of monensin/kg; $EC_{10} = 2.47$ mg of monensin/kg). The PNEC of this study can be calculated to be 247 µg/kg, applying an uncertainty factor of 10 (Tier B) to the EC_{10} of 2.47 mg/kg.

The updated risk ratios (< 1), reported in Table 2 using the same PEC_{soil} values calculated in the previous opinions (EFSA FEEDAP Panel, 2017, 2018), indicate that monensin from Monimax® does not pose a risk to terrestrial plants when used as coccidiostat in feed for chickens or turkeys for fattening according to the conditions of use established by the applicant.

Table 2: Risk characterisation (PEC/PNEC ratio) for the terrestrial compartment (plants) for monensin sodium

Species	PEC_{soil} refined (µg/kg)	EC_{10} (µg/kg)	PNEC ($EC_{10}/10$) (µg/kg)	Risk characterisation ratio (PEC/PNEC)
Turkeys for fattening	70	2,470	247	0.28
Chickens for fattening	78.0	2,470	247	0.32

PEC: predicted environmental concentration; PNEC: predicted no effect concentration; EC_{10} : concentration of the test substance that results in 50% of the exposed animals being adversely affected (i.e. both mortality and sub-lethal effects).

Based on the above results, the previous conclusions of the FEEDAP Panel on the risk assessment of monensin sodium from Monimax® (EFSA FEEDAP Panel, 2017, 2018) can be updated as follows: the use of monensin sodium from Monimax® in complete feed for chickens for fattening, chickens reared for laying and turkeys for fattening does not pose a risk for the aquatic compartment, the terrestrial compartment and sediment. The bioaccumulation potential of monensin in the environment is low.

3.1.2. 4,4'-Dinitrocarbanilide

The FEEDAP Panel previous assessments on the environmental risk of DNC for the terrestrial compartment and sediment have been updated in the following chapters considering the new data available. The conclusions are updated for the assessment done in chickens for fattening (representing the worst-case) and then extended to chickens reared for laying and turkeys for fattening.

3.1.2.1. Physico-chemical properties/fate and behaviour studies

The physicochemical properties of DNC were described in a previous scientific opinion (EFSA FEEDAP Panel, 2017) and are summarised in Table 3.

Table 3: Physicochemical properties of DNC

Property	Value	Unit
Octanol/water partition coefficient ($\log K_{ow}$) ⁽¹⁾	3.25 (pH 5) 3.21 (pH 7) 3.23 (pH 9)	–
Water solubility	0.0209 (pH 5–9, $20 \pm 0.5^\circ\text{C}$)	mg/L
Dissociation constant (pK_a) ⁽²⁾	12.44 ± 0.70 ⁽³⁾	–
Vapour pressure ⁽¹⁾	3.1×10^{-10}	Pa

–: no unit.

(1): Technical dossier FAD-2012-0032/Supplementary information June 2015/Annex 9.

(2): Technical dossier FAD-2012-0032/Supplementary information June 2015/Annex 3.

(3): Estimated value since the substance exhibits insufficient water solubility and ultraviolet-visible absorptivity to enable experimental determination.

3.1.2.2. DNC accumulation in the terrestrial compartment

Predicted environmental concentration in soil

Studies assessing DNC adsorption/desorption and biodegradation in soil were assessed by the FEEDAP Panel in its previous opinions and for DNC a K_{oc} of 74,128 was used for the assessment. In the same opinions, the FEEDAP Panel established a conservative time to degradation to 50% of the original concentration (DT_{50}) in soil of 1,191 days for DNC. This result was obtained from the available soil degradation study (OECD guideline 307 but with an extended incubation period up to 400 days at $20 \pm 2^\circ\text{C}$) where a biphasic soil degradation kinetics was observed;⁶ the DT_{50} was derived from the slower phase and then adjusted to a temperature of 12°C . The resulting DT_{50} was of 1,191 days at 12°C .

In its opinion on the safety and efficacy of Monimax for chickens for fattening, the FEEDAP Panel used a PEC_{soil} refined for persistent compound of $966.5 \mu\text{g}/\text{kg}$. This value was calculated based on the DT_{50} of 1,191 days (after 39 years of continual Monimax® usage for chickens for fattening) considering an annual application rate ($PEC_{soil \text{ initial}}$) of $185 \mu\text{g}/\text{kg}$.⁷

The FEEDAP Panel notes that the $PEC_{soil \text{ plateau}}$ value has to be considered the worst-case PEC_{soil} for DNC since several conservative assumptions have been used to derive it:

- 1) A total residue approach was assumed: no metabolism of DNC in chicken and no dissipation of DNC during the storage of chicken litter on farm were considered.
- 2) In laboratory experiments, DNC demonstrated biphasic degradation with about 50% of the applied dose degraded after at least 180 days in four different soils. $PEC_{soil \text{ plateau}}$ only considers the degradation of DNC occurring in the slow phase, disregarding any initial degradation.
- 3) The $PEC_{soil \text{ plateau}}$ evaluates the accumulation of DNC in soil without considering any further dissipation route.

Whilst there may be some uncertainty about the degradation of DNC, the established $PEC_{soil \text{ plateau}}$ ($966.5 \mu\text{g}/\text{kg}$) can reasonably be considered conservative and a suitable worst-case to be used for risk assessment.

Ecotoxicity studies terrestrial compartment and updated risk characterisation (PEC/PNEC ratios)

The applicant provided a study on the effects on soil microorganisms,⁸ on soil invertebrates (earthworms)⁹ and on plants¹⁰ that were already submitted and assessed in the previous applications (EFSA FEEDAP Panel, 2017, 2018). The FEEDAP Panel finds no reason to change the previous conclusions. The PEC/PNEC ratios have been recalculated for chickens for fattening considering the $PEC_{soil \text{ plateau}}$ of $966.5 \mu\text{g}/\text{kg}$. Based on the new calculation, DNC is not expected to pose a risk for nitrogen transforming soil microorganisms. As regards earthworms, the $NOAEC_{\text{reproduction}}$ for DNC was $300,000 \mu\text{g}/\text{kg}$ and applying an assessment factor of 10, the PNEC of $30,000 \mu\text{g}/\text{kg}$ results in a PEC/PNEC ratio < 1 ($967/30,000 = 0.03$). In reference to plants, the EC_{50} for seedling and emergence was $> 248,000 \mu\text{g}/\text{kg}$ and applying an assessment factor of 100 resulted in a PNEC of $2,480 \mu\text{g}/\text{kg}$ and a PEC/PNEC ratio < 1 ($967/2,480 = 0.4$). The PEC/PNEC ratios are in line with those used in the previous FEEDAP assessment (EFSA FEEDAP Panel, 2018) and are < 1 .

Experimental data on DNC concentration in soils after repeated administration of poultry litter and model to estimate PEC_{soil}

In order to address the concerns expressed in the previous FEEDAP opinions ('DNC refined PECs showed uncertainties linked to the very high persistence of the compound'), the applicant provided also new data on the concentration of DNC in soils treated with poultry manure.¹¹ Nine agricultural fields in the EU (one in Belgium, four in Italy, one in Germany, two in the United Kingdom and one in Poland) were identified that had been fertilised with poultry litter fed nicarbazin-based feed additives for 4–10 years. Data on the potential total application of nicarbazin/DNC to those fields were based on information provided by poultry integrations or, when not available, estimated on the required feed additive usage on the farm. A number of soil samples were collected from January to February 2018 and analysed for their DNC content. The fields were estimated to receive 0.076–2.72 kg DNC/ha per year (Table 4).

⁶ Technical dossier/Reference 7 Sinclair 1.

⁷ Technical dossier/Review of EFSA environment safety/Table 1 of appendix.

⁸ Technical dossier/Reference 8 Sinclair 2.

⁹ Technical dossier/Reference 1 Arach 2011.

¹⁰ Technical dossier/Reference 9 Jarratt 1.

¹¹ Technical dossier/DNC soil persistence report.

Table 4: Characteristics of the fields selected for measuring DNC concentration in soil

Country	Field size (ha)	Years of ncarbazine application	Kg of feed additive applied ⁽¹⁾	Kg of ncarbazine applied ⁽²⁾	Kg of DNC applied ⁽³⁾	Kg/ha per year of DNC applied
Belgium	8	4	113.6	9.1	6.5	0.204
Italy (A)	45	5	301	24.1	17.1	0.076
Italy (B)	7	5	118	9.4	6.7	0.191
Italy (C)	15	5	774	61.9	43.9	0.585
Italy (D)	20	5	542	43.4	30.8	0.308
Germany	1.5	10	719	57.5	40.8	2.720
UK (A)	2.7	9	251.5	20.1	14.2	0.585
UK (B)	2.2	9	204.2	16.3	11.6	0.586
Poland	4	4	108	8.6	6.1	0.383

DNC: 4,4'-dinitrocarbanilide.

(1): Feed additive product usage estimation during the years indicated for each soil, based on the consumption per bird, birds treated and productive cycles per year.

(2): Based on ncarbazine composition of feed additive product of 80 g/kg

(3): Based on the molecular weight of DNC of 302.24 g/mol and HPD of 124.14 g/mol.

Ten samples (11 in the Polish soil), taken on two diagonals across the field, were collected from each field. To avoid edge effects, soil samples were not taken within 5 m of field edges and had a maximum depth of 30 cm. Soil extracts and calibration standards were analysed by liquid chromatography–tandem mass spectrometry (LC–MS/MS) method. The measured concentrations of DNC in soil for all nine sites ranged between 0.1 and 35.2 µg/kg dw (Table 5), resulting in much lower concentrations than the $PEC_{soil\ plateau}$ values estimated.

Table 5: Concentration of DNC in the different field soils sampled (µg/kg on dry matter basis)

Country	No. samples	Mean	Median	Standard deviation	Minimum	Maximum
Belgium	10	18.5	17.9	12.2	4.9	35.2
Italy (A)	10	7.1	5.0	4.9	2.9	17.6
Italy (B)	10	3.7	1.9	6.8	0.6	23.1
Italy (C)	10	0.7	0.6	0.3	0.3	1.1
Italy (D)	10	11.0	9.8	7.2	2.4	22.4
Germany	10	2.3	2.4	0.9	1.3	3.4
UK (A)	10	0.4	0.3	0.4	0.1	1.5
UK (B)	10	1.1	0.9	0.6	0.4	2.1
Poland	11	2.7	0.6	5.1	0.1	17.3

The measured DNC concentrations in soil were also used to build a simple model to estimate PEC_{soil} . The Belgian field (highest mean DNC concentration detected) was selected for modelling.

The following assumptions were made:

- DNC soil concentration prior to the start of applications was considered zero;
- No metabolism of the DNC in poultry and/or degradation during the litter storage on farm was considered;
- The field site received ncarbazine (DNC) containing poultry litter on four successive years (2014–2017) and the total amount of ncarbazine (DNC) was evenly distributed in each year;
- The application of poultry litter in each year occurred in one spreading event on last day of September (e.g. after the harvest of the crop and sowing of the following crop);
- The poultry litter and hence DNC is applied homogeneously across the field;
- The mixing depth in soil (assuming the sites had been ploughed) is 30 cm;
- Two DNC soil degradation rates from the above-mentioned report were considered,¹² both adjusted to 12°C: a slow rate degradation DT_{50} of 1,497.9 days (i.e. 700.5 days at 20°C); and an overall degradation DT_{50} of 206.6 days (i.e. 96.6 days at 20°C).

¹² Technical report/Reference 7 Sinclair 1.

The highest field measured soil concentration (35.3 µg/kg, Belgian site) was lower than the highest value modelled both assuming the highest DT₅₀ of 1,497.9 days (PEC_{soil} = 128.4 µg/kg) and the lowest DT₅₀ of 206.6 days (PEC_{soil} = 43.9 µg/kg). The differences could be attributable to the conservative assumptions made (no dissipation from metabolism and/or manure degradation) and/or with field soil degradation rates that are faster than those evaluated in laboratory with the soil degradation studies.

Conclusion

Based on the above results, the previous conclusions of the FEEDAP Panel on the risk assessment of DNC from Monimax® (EFSA FEEDAP Panel, 2017, 2018) can be updated as follows: the use of DNC from Monimax® in complete feed for chickens for fattening, chickens reared for laying and turkeys for fattening does not pose a risk for the terrestrial compartment.

3.1.2.3. DNC accumulation in the sediment compartment

In the previous opinions (EFSA FEEDAP Panel, 2017, 2018), a PEC_{sediment} for DNC was established using the output from the worst-case FOCUS surface water modelling scenario (R3) multiplied by a factor of 6.5 to account for 20 years of application (PEC_{sed} > 313 µg/kg for turkeys for fattening and > 351 µg/kg for chickens for fattening). This value was not considered a definitive value since a possible accumulation of DNC in sediment over successive years was considered and no data on the transformation of DNC in sediment were available.

A cumulative worst-case PEC_{sediment} concentration has been quantified by the applicant taking into account the accumulation of DNC in sediment over a 50-year period of continual DNC application following the use of Monimax® in chickens for fattening, to the same area of land and subsequent movement to the freshwater sediment compartment. As described in Section 3.1.2.2, PEC_{soil plateau} calculated using a worst-case soil DT₅₀ value of 1,191 days (at 12°C) was reached in 39 years. As a worst-case scenario, it was assumed that no degradation in sediment occurred over the 50-year accumulation period (DT₅₀ for sediment = 1,000,000 days). For each year, FOCUS surface water modelling for winter cereals has been performed for the R3 Stream scenario (the worst case one for DNC) with the following input parameters:

- DNC property: DT₅₀ soil 562 days (at 20°C); DT₅₀ sediment 1,000,000 days; DT₅₀ surface water 1,000 days (default); K_{oc} 74,128 L/kg (≈ K_{om} 42,998 L/kg); 1/n 0.9; molecular weight 302.24 g/mol; vapour pressure 3 × 10⁻¹⁰ Pa at 25°C; water solubility 0.0209 mg/L.
- Application: chemical application method soil incorporated, 4; uniform incorporation in soil uniform; depth incorporated 20 cm; application window 2 November to 2 December.

Each year the 'application rate' has been calculated summing the PEC_{soil} of the DNC remaining from the previous application and the PEC_{soil} of DNC from the new application. DNC containing litter was considered applied to land in a single spreading event each year with an annual addition to the soil of 185 µg/kg (chickens for fattening) and incorporated into soil at a depth of 20 cm. After 50 years continual application, the PEC_{sediment} was 15,409.5 µg/kg.

Risk to the sediment compartment incorporating degradation in sediment

The applicant provided also a new study on aerobic and anaerobic transformation in aquatic sediment systems, performed in accordance with OECD Guideline 308 to determine the rate and route of degradation of DNC in freshwater sediments.¹³ Two freshwater water-sediment systems (Nesthauser See Lake and Biggese Lake) with differing properties were treated with [DNC-phenyl-U-¹⁴C]-nicarbazine at a rate of 1,000 µg/kg sediment dw, applied to the water compartment, and maintained in the dark at 20 ± 2°C for up to 99 days. The Nesthauser Lake sediment had a low organic carbon content (0.5–2.5%) and a coarse texture and the Biggese Lake sediment had a high organic carbon content (2.5–7.5%) and a fine texture.

No metabolites were formed at levels greater than 10% of the applied radioactivity in both systems.

The peak concentration in the Nesthauser See Lake sediment was observed at 7 days after application; the corresponding DT₅₀ of 32.82 days (DT₉₀ = 113.2 days) at 20°C was obtained through single-first order kinetics (best fit). The peak concentration in the Biggese Lake sediment was observed at 14 days after application; the corresponding DT₅₀ of 71.42 days (DT₉₀ = 100.3 days) at 20°C was obtained through bi-phasic kinetic model 'hockey-stick' (best fit). The bi-phasic rate constants were 0.006594 day⁻¹ (k₁) and 0.103 day⁻¹ (k₂). The most conservative sediment DT₅₀ from

¹³ Technical dossier/Reference 6 Peck final.

the study would therefore be the one generated from the slowest rate constant observed (i.e. k_1 rate) from the degradation in Nesthauser See Lake sediment and would be 105.1 days (i.e. $\ln(2)/0.006594$) and the DT_{90} sediment would be 349.2 days (i.e. $\ln(10)/0.006594$) at 20°C (224.7 days and 746.7 days for DT_{50} and DT_{90} at 12°C, respectively).

The modelling described earlier was repeated using the same input values for all parameters, while the sediment DT_{50} was refined from 1,000,000 days (effectively no degradation) to the more realistic value of 105.1 days at 20°C generated from the study described above.

A DT_{50} of 224.7 days (at 12°C) was used to refine the DNC concentration remaining in the sediment from previous years applications for the current year. After 50 years continual application the PEC_{sediment} was 337.3 µg/kg, far lower than the previous calculation.

Updated risk characterisation (PEC/PNEC ratios)

In the previous opinion (EFSA FEEDAP Panel, 2017, 2018), a $PNEC_{\text{sediment}}$ of 24,100 µg/kg (dw) had been calculated from a *Chironomus riparius* study where a NOEC for the most sensitive endpoint was established at 241 mg/kg dw.¹⁴

The risk characterisation of DNC through the continual use of Monimax® for chickens for fattening, recalculated considering a 50-year period of use, results in a PEC/PNEC ratio < 1, indicating no risk to sediment-dwelling organisms, even when considering a cumulative worst-case PEC_{sediment} of 15,409.5 µg/kg ($15,409.5/24,100 = 0.64$).

Conclusion

Based on the above evidence, the previous conclusions of the FEEDAP Panel on the risk assessment of DNC from Monimax® (EFSA FEEDAP Panel, 2017, 2018) can be updated as follows: the use of DNC from Monimax® in complete feed for chickens for fattening, chickens reared for laying and turkeys for fattening does not pose a risk for sediment.

3.1.2.4. DNC potential for bioaccumulation and for secondary poisoning

In the previous scientific opinions (EFSA FEEDAP Panel, 2017, 2018), no data on bioaccumulation had been submitted and the FEEDAP Panel concluded that the high persistence and hydrophobicity of DNC ($\log K_{ow} > 3$) indicate that there might be a risk for bioaccumulation.

As regards the potential of DNC to bioaccumulate, the applicant submitted in-silico data estimating the DNC bioaccumulation factor (BCF) according to Cotterill and Ridgeway (2018).¹⁵ The value range of different models were between 8 and 140 L/kg, far below the trigger value for a compound to be bioaccumulative (> 2,000 L/kg).

In order to assess the risk of secondary poisoning, the method according to ECHA guidance has been considered (ECHA, 2008, 2016).¹⁵ Based on the data presented in the previous scientific opinion (EFSA FEEDAP Panel, 2017), the lowest no observed adverse effect level (NOAEL) for DNC, derived from 52-week chronic oral study in rat was 20 mg/kg body weight (bw) per day. Using a conversion factor of 20 (*Rattus norvegicus*) a NOEC value of 400 mg/kg feed was calculated. Based on these data and applying an assessment factor of 30, $PNEC_{\text{oral}}$ for DNC is 13.3 mg/kg feed ($400/30 = 13.3$). This value is higher than the estimated PEC_{oral} in worms and fish of 0.10 and 0.019 mg/kg, respectively (see Table 6). The PEC/PNEC ratios for surface water and soil are below 1. A risk for secondary poisoning for worm/fish eating birds and mammals is not likely to occur for DNC.

Table 6: Risk of secondary poisoning for DNC

	$PEC_{\text{oral, sw}}$ (mg/kg)	$PEC_{\text{oral, soil}}$ (mg/kg)	$PNEC_{\text{oral}}$ (mg/kg)	$PEC/PNEC_{\text{sw}}$	$PEC/PNEC_{\text{soil}}$
DNC	0.019	0.10	13.3	0.001	0.008

DNC: 4,4'-dinitrocarbanilide; PEC: predicted environmental concentration; PNEC: predicted no effect concentration.

Conclusion

Based on the above results, the previous conclusions of the FEEDAP Panel on the risk assessment of DNC from Monimax® (EFSA FEEDAP Panel, 2017, 2018) can be updated as follows: DNC does not bioaccumulate in the environment and the risk for secondary poisoning is not likely to occur.

¹⁴ Technical dossier/Reference 5 Jarrat 2.

¹⁵ Technical dossier/Review of EFSA environment.

3.2. Post-marketing monitoring

In the previous opinion (EFSA FEEDAP Panel, 2017), it was recommended to investigate the potential of DNC to accumulate in soil over the years by monitoring and a field study. The applicant already provided results from a field survey, showing that no concern for accumulation has to be expected.

The applicant proposes a monitoring plan that, in the light of the evidence reported above, is no more considered crucial.¹⁶

4. Conclusions

On the basis of the new data provided, the FEEDAP Panel updates as follow its previous conclusions on the safety of Monimax® for the environment:

The use of monensin sodium from Monimax® in complete feed for chickens for fattening, chickens reared for laying and turkeys for fattening poses no risk for the aquatic and terrestrial compartments or for sediment. The bioaccumulation potential of monensin in the environment is low.

No concerns would arise for the HDP moiety of nicarbazin excreted from chickens for fattening, chickens reared for laying and turkeys fed Monimax®. The use of DNC moiety of nicarbazin from Monimax® in complete feed for chickens for fattening, chickens reared for laying and turkeys for fattening poses no risk for the aquatic and terrestrial compartments or for sediment. The bioaccumulation potential of DNC in the environment is low and the risk for secondary poisoning is not likely to occur.

Overall, the additive Monimax®, when used in complete feed for chickens for fattening, chickens reared for laying and turkeys for fattening according to the conditions of use, poses no risk for the environment.

Chronology of the application of Monimax® for turkeys for fattening

Date	Event
22/11/2018 and 04/12/2018	Dossier received by EFSA: Monimax® (monensin sodium and nicarbazin) for turkeys for fattening. Submitted by Huvepharma NV
15/11/2018	Reception mandate from the European Commission
07/12/2018	Application validated by EFSA – Start of the scientific assessment
11/04/2019	Request of supplementary information to the applicant in line with Article 8(1)(2) of Regulation (EC) No 1831/2003 – Scientific assessment suspended. <i>Issues: safety for the environment</i>
06/05/2019	Reception of supplementary information from the applicant - Scientific assessment re-started
07/10/2019	Opinion adopted by the FEEDAP Panel. End of the Scientific assessment

Chronology of the application of Monimax® for chickens for fattening and chickens reared for laying

Date	Event
22/11/2018 and 04/12/2018	Dossier received by EFSA: Monimax® (monensin sodium and nicarbazin) for turkeys for fattening. Submitted by Huvepharma NV
12/04/2019	Reception mandate from the European Commission
16/04/2019	Application validated by EFSA – Start of the scientific assessment
07/10/2019	Opinion adopted by the FEEDAP Panel. End of the Scientific assessment

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Abbreviations

BCF	bioaccumulation factor
bw	body weight
DNC	4,4'-dinitrocarbanilide
DT ₅₀	time to degradation of 50% of original concentration of the compound in the tested soils
EC ₅₀	concentration of the test substance that results in 50% of the exposed animals being adversely affected (i.e. both mortality and sublethal effects).
ECHA	European Chemicals Agency
EMA	European Medicines Agency
FOCUS	Forum for coordination of pesticide fate models and their use
HDP	4,6-dimethylpyrimidin-2-ol
K _{oc}	organic carbon–water partitioning coefficient
K _{om}	organic matter/water distribution coefficient (L/kg). It corresponds to K _{oc} /1.724
LC–MS/MS	liquid chromatography–tandem mass spectrometry
Log K _{ow}	logarithm of octanol-water partition coefficient
MW	molecular weight
NOAEL	no observed adverse effect level
PEC	predicted environmental concentration
PNEC	predicted no effect concentration
UF	uncertainty factor