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SCIENTIFIC OPINION

Scientific Opinion on the safety and efficacy of formic acid, ammonium formate and sodium formate as feed hygiene agents for all animal species[1]

EFSA Panel on Additives and Products or Substances used in Animal Feed (FEEDAP)[2,3]

European Food Safety Authority (EFSA), Parma, Italy

ABSTRACT

This opinion concerns the authorisation for a new use of formic acid, ammonium formate and sodium formate used as feed hygiene agents for all animal species. Studies performed with formic acid or its salts are considered equivalent when these agents are used on an equimolar basis. Conclusions of the previous opinion on formic acid and its safety for target species, consumers and the environment are reiterated. No adverse effects are anticipated when formic acid is used at the maximum proposed dose (pigs 12 000 mg/kg, all other animal species 10 000 mg/kg formic acid equivalents/kg complete feed). For ammonium formate, the inevitable presence of formamide is considered insufficient to guarantee the protection of reproduction animals from developmental toxicity. Evidence of carcinogenic potential argues for avoiding its use in reproducing animals and non-food-producing animals. The use of formic acid and sodium formate in animal nutrition is safe for consumers. Use of ammonium formate in dairy animals and laying poultry gives rise to concerns because of the potential exposure of consumers to formamide. Formic acid and its salts are corrosive and skin sensitisers. Sodium formate is mildly irritating to the eyes. Ammonium formate is considered an irritant for skin and eyes. The exposure via inhalation is considered to present a risk to unprotected workers handling the additive. The use of formic acid and its salts in animal nutrition is safe for the environment. Formic acid, at recommended concentrations, is effective at inhibiting or reducing the numbers of bacterial pathogens in feed, fulfilling the classical requirements of a preservative additive. Limited data are available to demonstrate the effects of formate salts in feed. Decreasing the number of viable microbial cells in contaminated feed does not eliminate the potential hazards associated with bacterial toxins and endotoxins that may be present in feed.

KEY WORDS

formic acid, ammonium formate, formamide, sodium formate, feed hygiene agent, safety, efficacy

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SUMMARY

Following a request from the European Commission, the Panel on Additives and Products or Substances used in Animal Feed (FEEDAP) was asked to deliver a scientific opinion on the application for the authorisation of formic acid, ammonium formate and sodium formate when used as feed hygiene agents for all animal species.

Formic acid, ammonium formate and sodium formate are currently authorised for use as preservatives in feed for all animal species.

Formate is a natural constituent of ruminal and intestinal contents and manure. Since animals are exposed to formate, studies performed with formic acid or its salts are considered equivalent when these agents are used on an equimolar basis.

The FEEDAP Panel is not aware of any new information that would lead to a revision and, therefore, it reiterates the previous conclusions of the opinion on formic acid and its safety for target species, consumers and the environment.

No adverse effects are anticipated when formic acid and sodium formate are used at the maximum proposed dose in feed for pigs (12 000 mg formic acid equivalents/kg complete feed), poultry or ruminants (10 000 mg formic acid equivalents/kg complete feed). A margin of safety could not be identified. These conclusions are extrapolated to other animal species provided the maximum dose applied does not exceed 10 000 mg formic acid equivalents/kg complete feed.

Although the same conclusion would apply to ammonium formate itself, the inevitable presence of formamide as a contaminant in ammonium formate additives is of concern. Even if the concentration of formamide in the additive is restricted to a maximum of 0.3 %, as proposed, this is considered insufficient to guarantee the protection of reproduction animals from developmental toxicity. In addition, there is evidence from animal studies of a carcinogenic potential. These observations indicate that the use of ammonium formate-containing additives in reproducing animals and non-food-producing animals should be avoided.

The use of formic acid, ammonium formate or sodium formate in animal nutrition is not expected to contribute to consumer exposure to formate. The FEEDAP Panel concludes that the use of formic acid and sodium formate in animal nutrition is safe for the consumer. Although the use of ammonium formate in growing/fattening animals would not give rise to any safety concerns for the consumer, its use in dairy animals and laying poultry may give rise to concerns because of the potential exposure of consumers to formamide.

Formic acid, ammonium formate and sodium formate are corrosive. Sodium formate is non-irritating to the skin but mildly irritating to the eyes. In the absence of relevant data, ammonium formate is considered an irritant for skin and eyes. In the absence of relevant data, the three compounds should be considered skin sensitisers. The free acid is volatile, and exposure via inhalation is considered to present a risk to unprotected workers handling the additive. It is prudent to consider ammonium and sodium formate as respiratory irritants.

The use of formic acid, ammonium formate and sodium formate in animal nutrition is safe for the environment.

Formic acid, at recommended concentrations, could be effective in inhibiting or reducing the numbers of bacterial pathogens in feed materials and compound feeds. For ammonium formate and sodium formate, limited data are available to demonstrate their effects in feed. Efficacy effects seen for formic acid fulfil the classical requirements of a preservative additive.

The FEEDAP Panel notes that decreasing the number of viable microbial cells in contaminated feed does not eliminate the potential hazards associated with bacterial toxins and endotoxins that may be present in feed.

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BACKGROUND

Regulation (EC) No 1831/2003[4] establishes the rules governing the Community authorisation of additives for use in animal nutrition. In particular, Article 4(1) of that Regulation lays down that any person seeking authorisation for a feed additive or for a new use of a feed additive shall submit an application in accordance with Article 7.

The European Commission received a request from FEFANA/HYFAC[5] for authorisation of the products formic acid, ammonium formate and sodium formate when used as feed additives for all animal species (category: technological additive; functional group, new: feed hygiene agent) under the conditions mentioned in Table 1.

According to Article 7(1) of Regulation (EC) No 1831/2003, the Commission forwarded the application to the European Food Safety Authority (EFSA) as an application under Article 4(1) (authorisation of a feed additive or new use of a feed additive) taking in to consideration the new functional group proposed. EFSA received directly from the applicant the technical dossier in support of this application.[6] According to Article 8 of that Regulation, EFSA, after verifying the particulars and documents submitted by the applicant, shall undertake an assessment in order to determine whether the feed additive complies with the conditions laid down in Article 5. The particulars and documents in support of the application were considered valid by EFSA as of 21 January 2014.

Formic acid (E 236), ammonium formate (E 295) and sodium formate (E 237) are currently listed in the EU Register of Feed Additives as a technological additives (functional group: preservative) for use in feed for all animal species.[7]

They are also authorised in food legislation by Directive 2007/42/EC[8] as food packaging additive for use in "Ingredients for manufacturing" intended to come in contact with food. Formic acid is also authorised as food preservative in sauces and similar products (maximum 200 mg/kg) and water based flavoured drinks (maximum 100 mg/kg) (Codex, 2011).[9]

The Joint FAO/WHO Expert Committee on Food Additives established an acceptable daily intake (ADI) of formic acid of 0–3 mg/kg bw (JECFA, 1974; 1998).

The European Food Safety Authority (EFSA) issued two opinions on the safety and efficacy of formic acid and calcium formate as preservatives in feed for all animal species (EFSA FEEDAP Panel, 2014a,b).

TERMS OF REFERENCE

According to Article 8 of Regulation (EC) No 1831/2003, EFSA shall determine whether the feed additive complies with the conditions laid down in Article 5. EFSA shall deliver an opinion on the safety for the target animals, the consumer, the user and the environment and the efficacy of formic acid, ammonium formate and sodium formate, when used under the conditions described in Table 1.

[4] Regulation (EC) No 1831/2003 of the European Parliament and of the Council of 22 September 2003 on additives for use in animal nutrition. OJ L 268, 18.10.2003, p. 29.
[5] FEFANA/HYFAC (EU Association of Specialty Feed Ingredients and their Mixtures/Hygiene of the Feed Chain Authorisation Consortium). Avenue Louise, 130A, Box 1, 1050 Brussels, Belgium. Companies: Kemira Oyj; Perstorp AB; Selko Feed Additives; Andres Pintaluba S.A., BASF SE; Anitox Ltd.
[6] EFSA Dossier reference: FAD-2013-0036.
[7] European Union Register of Feed Additives pursuant to Regulation (EC) No 1831/2003. Available at: http://ec.europa.eu/food/food/animalnutrition/feedadditives/comm_register_feed_additives_1831-03.pdf
[8] Commission Directive 2007/42/EC of 29 June 2007 relating to materials and articles made of regenerated cellulose film intended to come into contact with foodstuffs. OJ L172, 30.06.2007, p. 71.
[9] Technical dossier/Section II/Section II Refs/II.7 Codex 2011.

Table 1: Description and conditions of use of the additive as proposed by the applicant

Additive	Formic acid (E 236) Ammonium Formate (E 295) Sodium Formate (E 237)
Registration number/EC No/No (if appropriate)	E236, E295, E237
Category of additive	1. Technological additives
Functional group(s) of additive	NEW - Feed Hygiene Agent Substances, or when applicable, micro-organisms which control the microbial contamination (either by inhibiting or, reducing the growth of already present microorganisms or by limiting the potential for development of microorganisms on the feed) of pathogenic and zoonotic agents in feed materials and compound feeds.

Description			
Composition, description	Chemical formula	Purity criteria (if appropriate)	Method of analysis (if appropriate)
Formic acid – E 236	HCOOH	Min. 84.5% (liquid)	ISO2114
Ammonium formate – E295	$HCOONH_4$	Min. 35% (liquid)	HPLC method (validated and verified)
Sodium formate – E237	HCOONa	Min. 98% (solid) Min. 15% (liquid)	

Trade name (if appropriate)	Not appropriate
Name of the holder of authorisation (if appropriate)	Not appropriate

Conditions of use				
Species or category of animal	Maximum Age	Minimum content	Maximum content	Withdrawal period (if appropriate)
		mg of active substance/kg of complete feedingstuffs		
Formic acid - E 236 - All animal species and categories except pigs	-	-	10 000 expressed as formic acid	
Formic acid - E 236 - Pigs	-	-	12 000 expressed as formic acid	
Ammonium formate – E295 - All animal species and categories except pigs	-	-	10 000 expressed as formic acid	
Ammonium formate – E295 - Pigs	-	-	12 000 expressed as formic acid	
Sodium formate – E237 - All animal species and categories except pigs	-	-	10 000 expressed as formic acid	
Sodium formate – E237 - Pigs	-	-	12 000 expressed as formic acid	

Other provisions and additional requirements for the labelling	
Specific conditions or restrictions for use (if appropriate)	-
Specific conditions or restrictions for handling (if appropriate)	-
Post market monitoring (if appropriate)	-
Specific conditions for use in complementary feedingstuffs or water (if appropriate)	-

Maximum Residue Limit (MRL) (if appropriate)			
Marker residue	Species or category of animal	Target tissue(s) or food products	Maximum content in tissues
-	-	-	-

ASSESSMENT

This opinion is based on data provided by a consortium involved in the production/distribution of formic acid, ammonium formate and sodium formate. The Panel on Additives and Products or Substances used in Animal Feed (FEEDAP) has sought to use the data provided, together with data from other sources, to deliver an opinion.

1. Introduction

Formic acid is an intermediate in normal one-carbon metabolism in microorganisms, plants and animals, contributing to transmethylation reactions before, ultimately, being oxidised to carbon dioxide. It can be detected in various amounts in plants and animals and, in some plants and insects, provides a specialised defence against predation. Background levels of formic acid in fruit, honey, wine, roasted coffee, evaporated milk and in cheese range from 1 to 2 000 mg/kg, and levels between 2.7 and 87 mg/kg are reported in brandies produced from apples, pears, plums and apricots (HSDB, 2006; OECD, 2010).

Formic acid (E 236) is currently listed in the EU Register of Feed Additives as a technological additive (functional group: preservative) and as a sensory additive (functional group: flavouring compounds) for use in feed for all animal species.[10] It is also allowed for the processing of by-products of fish origin not intended for human consumption (Regulation (EC) No 93/2005).[11]

Although not formally authorised in the EU for food use, it is registered under the General Standards for Food Additives provisions of the Food and Agriculture Organization of the United Nations (FAO) as a food preservative in sauces and similar products, to a maximum of 200 mg/kg, and in flavoured drinks, to a maximum of 100 mg/kg (Codex, 2011).[12] The Joint FAO/WHO Expert Committee on Food Additives established an acceptable daily intake of formic acid of 0–3 mg/kg body weight (bw) (JECFA, 1974; 1998). Formic acid is authorised by Directive 2007/42/EC[13] for use in the production of regenerated cellulose food packaging intended for contact with food.

Ammonium formate (E 295) and sodium formate (E 237) are currently listed in the EU Register of Feed Additives as technological additives (functional group: preservative) for use in feed for all animal species without time limit.[14]

The current application is for the authorisation for a new use of formic acid, ammonium formate and sodium formate as technological additives used as feed hygiene agents in feed for all animal species.

The applicant requested the authorisation of formic acid, ammonium formate and sodium formate under the new (currently non-existent) functional group "feed hygiene agents/substances" belonging to the category "technological additives", with the following definition: "substances, or when applicable microorganisms, which control the microbial contamination (either by inhibiting or reducing the growth of already present microorganisms or by limiting the potential for development of microorganisms on the feed) of pathogenic and zoonotic agents in feed materials and compound feeds".

[10] European Union Register of Feed Additives pursuant to Regulation (EC) No 1831/2003. Available online: http://ec.europa.eu/food/food/animalnutrition/feedadditives/comm_register_feed_additives_1831-03.pdf

[11] Commission Regulation (EC) No 93/2005 of 19 January 2005 amending Regulation (EC) No 1774/2002 as regards processing of animal by-products of fish origin. OJ L19, 21.01.2005, p. 34.

[12] Technical dossier/Section II/Section II Refs/II.7 Codex 2011.

[13] Commission Directive 2007/42/EC of 29 June 2007 relating to materials and articles made of regenerated cellulose film intended to come into contact with foodstuffs. OJ L 172, 30.06.2007, p. 71.

[14] European Union Register of Feed Additives pursuant to Regulation (EC) No 1831/2003. Available online: http://ec.europa.eu/food/food/animalnutrition/feedadditives/comm_register_feed_additives_1831-03.pdf

2. Characterisation

2.1. Characterisation of the additive

2.1.1. Formic acid

Formic acid (methanoic acid, HCOOH, Chemical Abstracts Service (CAS) No 64-18-6, European Inventory of Existing Commercial Substances (EINECS) No 200-579-1) is a colourless liquid with a pungent odour and a molecular weight of 46.03 g/mol. Formic acid is soluble in water and alcohol, has a density of 1.22 g/cm^3 at 20 °C and a pK_a of 3.75 at 20 °C. Formic acid is a strong organic acid with corrosive properties.

Two commonly used methods of synthesis are described in the application. One method is based on the reaction of methanol and carbon monoxide, and the other on the hydrolysis of sodium formate. The products obtained from both methods are specified to contain a minimum of 84.5 % formic acid, the remainder being water.

Compliance with the specification was demonstrated by the analysis of five batches each from two companies (range: 84.8 to 85.7 % (w/w)).[15,16,17]

Commercial batches of formic acid were analysed for impurities.[18] The values obtained from three commercial batches, produced from carbon monoxide and methanol, for heavy metals (lead (Pb), cadmium (Cd) and mercury (Hg)), arsenic (As) and methanol met the specifications and were all below the limit of quantification (methanol < 500 mg/kg, methyl formate < 2 mg/kg, As < 0.10 mg/kg, Pb < 0.10 mg/kg, Cd < 0.10 mg/kg, Hg < 0.10 mg/kg). No data were provided for formic acid produced from sodium formate.

2.1.2. Ammonium formate/formic acid

Ammonium formate ($HCOONH_4$, CAS No 540-69-2, EINECS No 208-753-9) is a deliquescent crystalline solid with a molecular weight of 63.06 g/mol. Ammonium formate is soluble in water (143 g/L at 20 °C) and alcohol, and has a density of 1.26 g/cm^3.[19]

Ammonium formate can be produced by adding gaseous ammonia or an aqueous solution of ammonium hydroxide to formic acid. In both cases, there is only a partial neutralisation resulting in a mixture of the salt and the free acid. The additive is specified to contain a minimum of 35 % ammonium formate and a maximum of 64 % free formic acid.

Compliance with the specification was demonstrated by analysis of seven batches (four batches produced with ammonia gas and three batches produced with ammonium hydroxide). Independent of the production method, ammonium formate was in the range of 35.5 to 38.1 % (w/w). The amount of free formic acid varied depending on the production process. It was higher when gaseous ammonia was added (61.1 to 63.1 % (w/w)) than when ammonium hydroxide was used (24.8 to 25.3 % (w/w)). The water content also varied depending on the production process, being higher in the case of the ammonium hydroxide method (37.4 to 38.8 % (w/w)).[20,21]

Three commercial batches of ammonium formate/formic acid from each production method were analysed for impurities. The concentrations of heavy metals and arsenic were generally low and of no concern (ammonia gas method: Pb < 1 mg/kg, Cd < 0.1 mg/kg, Hg < 0.01 to < 0.1 mg/kg,

[15] Technical Dossier/Section II/Annex_II-1-1.
[16] Suplementary information (July 2014)/ Formic acid liquid-method 1-CoAs.
[17] Suplementary information (July 2014)/ Formic acid liquid-method 2-CoAs.
[18] Technical Dossier/Section II/Annex_II-1-1.
[19] Technical Dossier/Section II/ Sect_II_Identity.
[20] Technical Dossier/Section II/Annex_II-1-2a.
[21] Technical Dossier/Section II/Annex_II-1-2c.

As < 1 mg/kg; ammonium hydroxide method: Pb < 2 mg/kg; Cd < 0.2 mg/kg, Hg < 0.5 mg/kg, As < 0.2 mg/kg).

Formamide is inevitably generated in low concentrations during the production of ammonium formate. The applicant proposes a maximum level of 0.3 % w/w as the specification for formamide. Concentration of formamide in six batches of ammonium formate produced using ammonium hydroxide solution ranged between 0.08 and 0.27 % w/w, when measured after storage periods ranging from 1 to 19 months[22] (see Section 3.1.1).

2.1.3. Sodium formate (solid)

Sodium formate solid (formic acid, sodium salt; HCOONa, CAS No 141-53-7, EINECS No 205-488-0) is soluble in water and slightly soluble in alcohol, and has a density of 1.92 g/cm^3 at 20 °C.

Solid sodium formate is obtained as a by-product in the production of pentaerythritol, trimethylolpropane or neopentyl glycol. One of the starting materials (acetaldehyde, *n*-butyraldehyde or iso-butyraldehyde) is combined with formaldehyde and sodium hydroxide. The resulting sodium formate is isolated by crystallisation. The product is specified to contain ≥ 98 % sodium formate, the rest being water.

The analysis of five batches of sodium formate confirmed the specification (99.5 % sodium formate (w/w); range 99.4–99.7 %).[23]

Five commercial batches of solid sodium formate were analysed for impurities. The values obtained for heavy metals and arsenic were as follows: As < 1 mg/kg, Pb < 1 mg/kg, Cd < 0.1 mg/kg, Hg < 0.1 mg/kg.[24] The values of formaldehyde (0.3–6.2 mg/kg) from three batches were below the action limits set by the applicant (maximum of 10 mg/kg) and were not of concern.

Particles size distribution was determined by laser diffraction[25] for two batches of the additive, each prepared from a different process. Particles with diameters of less than 100 µm amounted to 23 and 50 % of each batch, and particles with diameters of less than 50 µm amounted to 9 and 19 % of each batch. The corresponding data for the respirable fraction (diameters of < 10 µm) were between 1 and 2.5 %. No data on dusting potential were provided.

2.1.4. Sodium formate/formic acid (liquid)

Sodium formate (for chemical description see section 2.1.3) has a molecular weight of 68.01 g/mol. The colourless liquid has a density of 1.30 g/cm^3 at 20 °C.

Liquid sodium formate is produced by mixing an aqueous solution of sodium hydroxide (50 % w/w) with formic acid. Formic acid is obtained by the manufacturing process described under section 2.1.3. The product is specified to contain a minimum of 15 % sodium formate, a maximum of 75 % free formic acid and a maximum of 25 % water.

The analysis of five batches showed mean contents of 20.2 % (w/w) sodium formate (range: 20.1 – 20.4 %), 62.0 % (w/w) formic acid (range: 61.7–62.2 %) and 17.8 % (w/w) water (range: 17.7–17.9 %).[26]

Three commercial batches were analysed for heavy metals and arsenic.[27] The values obtained for arsenic and heavy metals (As < 1 mg/kg, Pb < 1 mg/kg, Cd < 1 mg/kg, Hg < 0.1 mg/kg,

[22] Supplementary information (February 2015)/ Annex A.
[23] Supplementary information (July 2014)/ Na-formate solid CoAs.
[24] Technical Dossier/Section II/Annex_II-1-3b.
[25] Technical Dossier/Section II/Annex_II-1-4a.
[26] Technical Dossier/Section II/Annex_II-1-2b.
[27] Supplementary information (July 2014)/Impurities Na-formate-liquid.

iron < 3 mg/kg) were generally low and of no concern. No data were submitted for the formaldehyde content.

2.2. Stability and homogeneity

2.2.1. Shelf-life

No losses of formic acid (liquid) were observed after a three-year storage period at 20 or 25 ºC (three batches, in plastic bottles, recovery 99.8 %).[28]

Stability studies were performed with three batches of the liquid ammonium formate/formic acid preparation stored at 25 °C over a period of 24 months, and 40 °C over a period of six months.[29] The additive was stored in stainless steel or plastic containers. The results showed that the additive remained fully stable in plastic containers for the duration of the study but that losses amounting to approximately 10 % of the initial formate content were seen in the material stored in steel containers.

No significant losses of sodium formate (solid) were observed over a three-year period (three batches, recovery 99.2–99.8 % sodium formate) when stored under ambient conditions.[30] Three batches of sodium formate (liquid) stored for one year at 21–25 °C in plastic bottles showed no losses of sodium formate or formic acid.[31]

2.2.2. Stability in vitamin–mineral and acid premixtures

Typical layer, piglet and ruminant vitamin–mineral premixtures were formulated to contain about 15 % sodium formate solid. No losses of sodium formate were observed after six months' storage at ambient temperature.[32]

Formic acid and ammonium formate were added in different concentrations to four mixtures of other organic acids (lactic and acetic). All samples were stored at 25 °C for 12 months and were protected from light. No losses of formic acid or ammonium formate were observed (recovery: 95–105 %).[33]

In a separate study, formic acid (38 %) and sodium formate (23 %) were mixed with propionic acid. Three batches of this blend were stored at room temperature (21–24 °C) for 12 months. No losses of formic acid or sodium formate were observed.[34]

2.2.3. Stability in feedingstuffs

2.2.3.1. Formic acid

An additive containing 85 % formic acid was added at 7 000 mg/kg to a turkey feed in pelleted form and at 4 000 mg/kg to a pig feed in mashed form. Five samples of each were stored at room temperature for three months. In both feed types, no losses of formic acid were recorded.[35]

2.2.3.2. Ammonium formate/formic acid and sodium formate/formic acid

Both additives were added at a concentration of 10 000 mg/kg to a chicken, a piglet and a ruminant compound feed. The chicken and piglet diets were studied in mashed and pelleted form, and the ruminant feed only in pelleted form. Pelleting (at about 75 °C) did not affect the stability of the additives (difference by processing < 3.5 %). Storage for three months at ambient temperature resulted

[28] Technical Dossier/Section II/Annex_II-3-1.
[29] Technical Dossier/Section II/Annex_II-3-2.
[30] Supplementary information (July 2014)/Annex stability sodium formate solid.
[31] Technical Dossier/Section II/Annex_II-3-3.
[32] Technical Dossier/Section II/Annex_II-3-6.
[33] Technical Dossier/Section II/Annex_II-3-5a.
[34] Technical Dossier/Section II/Annex_II-3-5b.
[35] Technical Dossier/Section II/Annex_II-3-4.

in mean (mashed and pelleted) losses of 4.3 % for the ammonium formate and 3.7 % for the sodium formate.[36]

2.2.4. Homogeneity

2.2.4.1. Formic acid

Formic acid concentrations from the stability study with 10 samples each of two feed types were used to calculate coefficients of variation (CVs), which were 2 and 3 % for the turkey and the pig feeds, respectively, indicating the ability of the additive to distribute homogeneously in feedingstuffs.[37]

2.2.4.2. Ammonium formate/formic acid and sodium formate/formic acid

Formate concentrations from the stability study with 10 samples each of five feed types were used to calculate CVs. CVs of 2.3 % for both additives, measured as a mean of the broiler, piglet and ruminant feeds, indicate the ability of the additives to distribute homogeneously in feedingstuffs.[38]

2.3. Conditions of use

Formic acid and its ammonium and sodium salts are intended for use as feed hygiene agents in feed for all animal species without limitations by age. No withdrawal time is foreseen.

The proposed use levels for the additives are 10 000 mg/kg for pig feed (maximum content 12 000 mg/kg) and 5 000 mg/kg feed for all other species (maximum content 10 000 mg/kg feed), all expressed as formic acid/kg complete feed. Concentrations between 10 000 and 40 000 mg/kg feed may be used for individual feed materials.

2.4. Evaluation of the analytical methods by the European Union Reference Laboratory (EURL)

EFSA has verified the EURL reports as they relate to the methods used for the control of the formic acid, ammonium formate and sodium formate in animal feed. The Executive Summaries of the EURL reports can be found in Annex A and in Annex B.

3. Safety

Formic acid and its salts dissociate at physiological pH to formate, an important constituent of intermediary one-carbon metabolism. The safety assessment of formate is thus independent of whether the anion derives from the free acid or its salts. However, in the case of the salts, any impact of the accompanying cation is also taken into consideration. The safety implications of the presence of contaminating formamide present in the ammonium salt is also addressed.

3.1. Safety for the target species

In an opinion on the safety of formic acid (EFSA FEEDAP Panel, 2014a), the Panel concluded that "considering the results of the tolerance studies in chickens for fattening and cattle for fattening and of other published studies in which formic acid has been fed to target species at concentrations greater than that proposed by the applicant, no adverse effects are to be anticipated when formic acid is used at the maximum proposed dose in feed for pigs (12 000 mg formic acid/kg complete feed), poultry or ruminants (10 000 mg formic acid/kg complete feed). However, a margin of safety could not be identified. These conclusions are extrapolated to other animal species, provided the maximum dose applied does not exceed 10 000 mg formic acid/kg complete feed".

Since the same studies were made available for the purposes of this application, and as the Panel is unaware of any subsequent publication which would introduce any cause for concern, these

[36] Technical Dossier/Section II/Annex_II-3-6.
[37] Technical Dossier/Section II/Annex_II-3-4.
[38] Technical Dossier/Section II/Annex_II-3-6.

conclusions, as they relate to the formate ion, are taken to apply to the salts of formic acid under application.

No safety concerns arise from the presence of the cations occur, provided their concentration is taken into consideration when formulating diets.

3.1.1. Microbiological studies

As considered in previous opinions on formates (EFSA FEEDAP Panel, 2014a, 2015), microbiological studies are not considered necessary. Moreover, it is unlikely that formic acid or its formate salts present a risk of inducing cross-resistance to antibiotics and no data refuting this conclusion have been identified.

3.1.2. Safety of formamide

Formamide (CH_3NO, CAS No 75-12-7) is formed during the production of ammonium formate and/or during storage of the additive. When present as a contaminant of materials or mixtures, Regulation (EU) 1272/2008 (classification, labelling and packaging) requires labelling to indicate that formamide is categorised as (1) Reproductive toxicity; Category 1B; May damage fertility or the unborn child; and (2) Carcinogenicity; category 2; Suspected of causing cancer. For this reason, formamide or preparations containing it in concentrations equal to or higher than 0.3 % may not be placed on the EU market for sale to the general public. However, this regulation does not apply to food or feed or to food/feed additives. Formamide showed no evidence of mutagenicity in a series of relevant short-term bioassays (Ames, micronucleus (peripheral blood) and dominant lethal assays).[39] However, it did give a positive result in a micronucleus test with mouse bone marrow following intraperitoneal injection of 900 mg/kg bw or higher. This led to the conclusion that, although not mutagenic *in vitro*, formamide in high doses showed evidence of clastogenicity *in vivo* (OECD, 2007).

Oral gavage rat and mouse carcinogenicity studies, using formamide at 0, 20, 40 or 80 mg/kg bw, five days per week for 105 weeks, and following Organisation for Economic Co-operation and Development (OECD) Guideline 451 under Good Laboratory Practice, gave no evidence of a carcinogenic potential in male or female F344/N rats. However, there was evidence of carcinogenic activity of formamide in male B6C3F1 mice based on increased incidences of haemangiosarcoma of the liver. There was also evidence of carcinogenic activity of formamide in female B6C3F1 mice based on increased incidences of hepatocellular adenoma or carcinoma (combined), with the difference being significant at the highest dose. However, since incidence was within the historical range for the colony, this result was considered equivocal. Mineralisation of the testicular arteries and tunic and haematopoietic cell proliferation of the spleen in male mice were also associated with the administration of formamide.

Formamide was found to be embryotoxic and teratogenic in several oral gavage studies with rabbits, rats and mice. In mice treated during gestation, embryo toxicity and teratogenicity were seen in the absence of maternal toxicity. Mechanistic studies identified susceptible stages during gestation in both rats and mice. Among other animals, the rabbit was the most sensitive species to formamide in terms of developmental toxicity. The lowest No Observed Adverse Effect Level (NOAEL) identified (in rabbits) was 34 mg/kg bw for both maternal toxicity and embryo/fetotoxicity. In all studies, the identified NOAELs for teratogenicity were substantially higher.

The intake of formamide in reproduction animals given ammonium formate-containing products can be estimated using the default values applied by the Panel. For sows, the potential exposure can be calculated assuming a daily feed intake of 6 kg per sow and an inclusion of 2.3 % of the aqueous ammonium formate/formic acid additive delivering the maximum of 12 000 mg formate/kg feed with a maximum level of formamide contamination restricted to 0.3 %. Assuming a sow weight of 200 kg, this would mean an exposure of approximately 2.1 mg/kg bw per day. Equivalent calculations for

[39] Supplementary information (June 2013)/Annex B.

laying hens and dairy cows give estimated exposures of 3.4 and 1.8 mg/kg bw per day, respectively, similar to that of sows.

This difference between the potential exposure and the NOAEL identified in the rabbit study is considered insufficient to guarantee the protection of reproduction animals.

3.1.3. Conclusion on the safety for the target species

The FEEDAP Panel concludes that no adverse effects are to be anticipated when formic acid and sodium formate are used at the maximum proposed levels in complete feed for all animal species.

Although the same conclusion would apply to ammonium formate itself, the inevitable presence of formamide as a contaminant of ammonium formate additives is of concern. Even if the concentration of formamide in the additive is restricted to a maximum of 0.3 %, as proposed, this is considered insufficient to guarantee the protection of reproduction animals from developmental toxicity. In addition, there is evidence from animal studies of carcinogenic potential. These observations indicate that the use of ammonium formate-containing additives in reproducing animals and non-food-producing animals should be avoided.

3.2. Safety for the consumer

The FEEDAP Panel is not aware of any new information that would lead to a revision of the conclusions of the opinion on formic acid (EFSA FEEDAP Panel, 2014a) or of the opinion on ammonium, sodium and calcium formate (EFSA FEEDAP Panel, 2015) with regard to effects on the consumer, which are summarised below.

Salts of formic acid dissociate at physiological pH and the anion enters the one-carbon pool of the body or is further oxidised to carbon dioxide and water in the liver and the erythrocytes. Any residual unmetabolised formate is excreted via urine, faeces or expired air (Hanzlik et al., 2005).

Toxicokinetic studies in rats and mice have shown that formamide is readily absorbed after oral application, with maximum plasma concentrations seen after one to two hours. A substantial fraction is excreted unchanged in urine (30 %) and through degradation (CO_2: 30 % in rats and 50 % in mice), with only a minor amount seen in faeces (1–3 %). The half-life of formamide in plasma has been found to be 15 hours in rats and only 4–6 hours in mice (RTI, 1996, cited by OECD, 2007; MRI, 1998, cited by OECD, 2007).[40] These findings, as well as the fact that formamide is not lipid soluble, make it unlikely that residues of formamide would accumulate in animal tissues. No information was provided regarding the transfer of formamide to milk or eggs. The concentrations of related compounds, N,N-dimethyl formamide, N-hydroxymethyl-N-methylformamide and N-methylformamide, were found in the milk of rats at concentrations equal to those in the plasma of rats receiving oral N,N-dimethyl formamide (Saillenfait et al., 1997). It cannot be excluded that a fraction of the absorbed formamide may be transferred to milk and eggs and therefore consumers of dairy products and eggs could be exposed to formamide.

The use of formic acid, ammonium formate or sodium formate in animal nutrition is not expected to contribute to consumer exposure to formate. The FEEDAP Panel concludes that the use of formic acid and sodium formate in animal nutrition is safe for the consumer. Although the use of ammonium formate in growing/fattening animals would not give rise to any safety concerns for the consumer, its use in dairy animals and laying poultry may give rise to concerns because of the potential exposure of consumers to formamide.

[40] Supplementary information (June 2013)/Annex B.

3.3. Safety for the user

The ammonium and sodium salts of formic acid are less corrosive alternatives to formic acid. However, the liquid forms of the ammonium formate and sodium formate additives contain a considerable amount of formic acid; therefore, studies on formic acid are considered relevant.

3.3.1. Effects on skin and eyes

Formic acid is recognised to be corrosive to skin and eyes, and ammonium formate/formic acid and sodium formate/formic acid are assumed to have similar properties.

Sodium formate was examined for skin and eye irritancy following OECD Guideline 402[41] and EPA OTS (Environmental Protection Agency, Office of Toxic Substances) Guideline 798.4500,[42] respectively (OECD, 2010). Sodium formate was found to be non-irritating to skin but mildly irritating to the eyes. No dermal sensitisation tests were reported. In the absence of relevant data, ammonium formate is considered an irritant for skin and eyes.

3.3.2. Effects on the respiratory system

The free acid is volatile, and exposure via inhalation is considered to present a risk to unprotected workers handling the additive (OECD, 2010). This also applies to the liquid forms of ammonium and sodium formate which include a significant proportion of the free acid.

Mild respiratory irritation was also observed in a single study with pure sodium formate, which essentially followed the protocol for OECD Guideline 403 toxicity studies (OECD, 2010).[43] Minor adverse effects were seen, which disappeared one week after exposure. Since the particle size distribution data available indicate that 9–19 % of sodium formate contains particles with diameters of less than 50 μm, exposure via a respiratory route is possible. Accordingly, it would be prudent to consider sodium and ammonium formate as respiratory irritants.

3.4. Safety for the environment

The FEEDAP Panel is not aware of any new information that would lead to a revision of the conclusions of the opinion on formic acid (EFSA FEEDAP Panel, 2014a) or of the opinion on ammonium, sodium and calcium formate (EFSA FEEDAP Panel, 2015) with regard to effects on the environment, which are summarised below.

Formic acid supplied at the rate specified in these opinions will increase methane emissions per bovine by, on average, about 5 %. Given that methane emissions by ruminants constitute around 2 % of total greenhouse gas emissions, and that only a small proportion of ruminants would receive formate as an additive, it was concluded that the impact of supplemental formic acid or its salts on the environment would be minor. Therefore, the use of formic acid, ammonium formate and sodium formate in animal nutrition is safe for the environment.

4. Efficacy

The applicant requested the classification of formic acid, ammonium formate and sodium formate under a new (currently non-existent) functional group "feed hygiene agents/substances" under the category "technological additives", with the following definition: "substances, or when applicable microorganisms, which control the microbial contamination (either by inhibiting or reducing the growth of already present microorganisms or by limiting the potential for development of microorganisms on the feed) of pathogenic and zoonotic agents in feed materials and compound feeds".

[41] Technical Dossier/Section III/ Sect III Refs/III.77.
[42] Technical Dossier/Section III/ Sect III Refs/III.83.
[43] Technical Dossier/Section III/ Sect III Refs/III.74.

Regulation (EC) No 1831/2003 has already established, under the category "technological additives", the functional group "preservatives" for substances which limit microbial growth. Preservatives are defined as having the following function: "inhibition of microbial growth, particularly that of biotic and spoilage organisms".[44] The FEEDAP Panel's guidance on technological additives amends this definition to "inhibition of microbial growth, particularly that of known pathogenic and spoilage organisms".

The FEEDAP Panel notes that the differences between the functional group "preservatives" and the newly proposed functional group "feed hygiene agents/substances" appear to be marginal. Although the formulations "reduction of the growth of already present microorganisms" and "limiting the potential for development of microorganisms on the feed" may appear to be new elements in the definition of the functional group "feed hygiene agents/substances", the practice of the evaluation of efficacy of preservatives was already based on data demonstrating these effects. Therefore, the FEEDAP Panel applies the criteria already defined for preservatives to evaluate the efficacy of formic acid, ammonium formate and sodium formate as feed hygiene agents/substances.

4.1. Minimum inhibitory concentration of formic acid and its salts

The applicant provided several studies on the minimum inhibitory concentration (MIC) of formic acid and its ammonium and sodium salts for several pathogenic bacteria (Östling and Lindgreen, 1993; Richards et al., 1995; Strauss and Hayler, 2001; Chaveerach et al., 2003; Nakai and Siebert, 2003; Boyen et al., 2008).[45,46] The MICs were expressed in equivalent percentages by calculation from mmol/L. The data showed that formic acid can inhibit the growth of several bacterial strains, particularly *Salmonella* spp., *Campylobacter* spp. and *Listeria monocytogenes*, at calculated concentrations of between 0.006 and 1.3 %. The MICs increased with increasing pH of the media. Thus, the antimicrobial activity of formic acid and its salts was demonstrated in pure cultures. It is not possible, however, to extrapolate from these data to quantify efficacy in feeds or premixtures.

4.1.1. Artificially contaminated feed

4.1.1.1. Study 1

Compound feed, mashed and pelleted, containing 0 and 5 000 mg/kg formic acid was incubated with *Salmonella enterica* subsp. *enterica* serovar Enteritidis (1×10^6 colony-forming units (CFU)/g). Changes in the bacterial count and pH were recorded after 0, 6, 24, 72 and 144 hours. *S.* Enteritidis numbers decreased with time in all samples (including control). However, after 144 hours, about 4×10^2 CFU/g of *S.* Enteritidis were found in the untreated groups but less than 50 CFU/g were found in the formic acid feed samples.[47]

4.1.1.2. Study 2

A pelleted broiler feed was treated with 3 000, 6 000, 9 000 and 12 000 mg/kg of a commercial product containing 85 % formic acid. Feeds were inoculated with *S.* Enteritidis, *S.* Typhimurium and *Campylobacter jejuni* at levels of 1×10^6 CFU/g each, and 0.2×10^6 CFU/g of an enterobacteria mix (*Escherichia coli*, *Citrobacter freundii*, *Proteus mirabilis*, *Enterobacter cloacae* and *Serratia ficaria*; total 1×10^6 CFU/g). Reductions in the *Salmonella* and *Campylobacter* counts were recorded after 0, 6, 24, 72 and 144 hours in all samples. However, *Salmonella* decreased more rapidly and to a lower final concentration in treated samples (9 000 and 12 000 mg/kg). No effect was observed on the enterobacteria mix.[48]

[44] Commission Regulation (EC) No 429/2008 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, 22.5.2009, p. 64.
[45] Technical Dossier/Section IV/Sect IV Refs/IV.4.
[46] Technical Dossier/Section IV/Sect IV Refs/IV.5.
[47] Technical Dossier/Section IV/Sect IV Refs/IV.12.
[48] Technical Dossier/Section IV/Sect IV Refs/IV.14.

4.1.1.3. Study 3

Pig mash diet artificially contaminated with *S.* Typhimurium, *E. coli* or *C. jejuni* was treated with formic acid (85 %) or ammonium formate (88 % formic acid equivalent) at 0, 4 000, 6 000, 8 000 or 10 000 mg/kg. Bacterial counts performed 4, 24 and 48 hours after treatment were lower in all of the treated groups than in the control group.[49]

4.1.1.4. Study 4

Poultry feed artificially contaminated with *S.* Gallinarum (1.36×10^6 CFU/kg) was treated with 0, 5 000, 10 000 and 15 000 mg/kg formic acid (85 %) and stored for seven days. Significant reductions ($P < 0.05$) in viable numbers of *S.* Gallinarum were reported for all treatments in a dose- and time-dependent manner compared with the control (Al-Natour and Alshawabkeh, 2005).

4.1.2. Naturally contaminated feed

4.1.2.1. Study 1

Salmonella survival in naturally contaminated feed materials treated with formic acid was measured (Hansen et al., 1995). In a laboratory study, cottonseed and rapeseed expellers (250 CFU *Salmonella*/100 g; range: 30 to > 960 CFU *Salmonella*/100 g) were mixed with 10 000 or 40 000 mg/kg formic acid. Survival of *Salmonella* was less than 1 % in both feed materials, supplemented with formic acid at either concentration, after two days. In a pilot-scale study, compound feed showed a similar effect when treated with formic acid (15 000, 20 000 or 25 000 mg/kg) and stored for 14 days.

4.1.2.2. Study 2

Formic acid (2 000 mg/kg) was added to liquid feed under fermentation and incubated for 48 hours. Samples were taken after 0, 6, 24 and 48 hours. After 48 hours, 90 % of the mixture was removed and replaced with fresh feed and water, and samples were taken after 96, 102 and 108 hours of incubation. Lactic acid bacteria increased in the diet (from < 3.0 to 9.5 log CFU/g) and yeast counts were low (< 3.0 to 5.4 log CFU/g) during the entire incubation period. Enterobacteria counts decreased in the initial phase in the treated groups and in the final phase (96 to 108 hours) (Canibe et al., 2007).

4.1.2.3. Study 3

Salmonella contamination of breeder feed, breeder litter, papers on which chicks are taken to a broiler house (called insert papers) and hatchery waste (dead chicks) was significantly reduced ($P < 0.01$) after treatment of breeder feed with formic acid at concentrations of 5 000 ± 1 000 mg/kg complete feed. No significant effects were observed on the *Salmonella* content of caecal samples (Humphrey and Lanning, 1988).

4.2. Conclusion on efficacy

Formic acid at the recommended concentrations could be effective in inhibiting or reducing the numbers of bacterial pathogens in feed materials and compound feeds. It therefore fulfils the classical requirements of a preservative additive. Limited data are available to demonstrate the effects in feed of formate salts. The effects depend on the dose, the period between dosing and consumption, the type of bacterial pathogens and the feed quality (i.e. composition, moisture content and storage conditions).

The FEEDAP Panel notes that decreasing the number of viable microbial cells in contaminated feed does not eliminate the potential hazards associated with bacterial toxins and endotoxins that may be present in feed.

[49] Technical Dossier/Section IV/Sect IV Refs/IV.24.

CONCLUSIONS AND RECOMMENDATIONS

CONCLUSIONS

The FEEDAP Panel concludes that no adverse effects are to be anticipated when formic acid and sodium formate are used at the maximum proposed dose in feed for pigs (12 000 mg formic acid equivalents/kg complete feed), poultry or ruminants (10 000 mg formic acid equivalents/kg complete feed). A margin of safety could not be identified. These conclusions are extrapolated to other animal species provided the maximum dose applied does not exceed 10 000 mg formic acid equivalents/kg complete feed.

Although the same conclusion would apply to ammonium formate itself, the inevitable presence of formamide as a contaminant of ammonium formate additives is of concern. Even if the concentration of formamide in the additive is restricted to a maximum of 0.3 %, as proposed, this is considered insufficient to guarantee the protection of reproduction animals from developmental toxicity. In addition, there is evidence from animal studies of a carcinogenic potential. These observations indicate that the use of ammonium formate-containing additives in reproducing animals and non-food-producing animals should be avoided.

The use of formic acid, ammonium formate or sodium formate in animal nutrition is not expected to contribute to consumer exposure to formate. The FEEDAP Panel concludes that the use of formic acid and sodium formate in animal nutrition is safe for the consumer. Although the use of ammonium formate in growing/fattening animals would not give rise to any safety concerns for the consumer, its use in dairy animals and laying poultry may give rise to concerns because of the potential exposure of consumers to formamide.

Formic acid, ammonium formate and sodium formate are corrosive. Sodium formate is non-irritating to the skin but mildly irritating to the eyes. In the absence of relevant data, ammonium formate is considered an irritant for skin and eyes. In the absence of relevant data, the three compounds should be considered skin sensitisers. The free acid is volatile, and exposure via inhalation is considered to present a risk to unprotected workers handling the additive. It is prudent to consider ammonium and sodium formate as respiratory irritants.

The use of formic acid, ammonium formate and sodium formate in animal nutrition is safe for the environment.

Formic acid at recommended concentrations is effective in inhibiting or reducing the numbers of bacterial pathogens in feed materials and compound feeds. For ammonium formate and sodium formate, limited data are available to demonstrate their effects in feed. Efficacy effects seen for formic acid fulfil the classical requirements of a preservative additive.

The FEEDAP Panel notes that decreasing the number of viable microbial cells in contaminated feed does not eliminate the potential hazards associated with bacterial toxins and endotoxins that may be present in feed.

RECOMMENDATIONS

Ammonium formate/formic acid should not be stored in metal containers.

It should be mentioned under other provisions that the contribution of sodium formate to the sodium supply of animals should be considered when formulating diets.

The contents of formic acid equivalents in the additives under application should be specified, as well as the sodium content of sodium formate preparations.

DOCUMENTATION PROVIDED TO EFSA

1. Formic acid (E 236), Ammonium Formate (E 295), Sodium Formate (E 237) for all animal species. September 2013. Submitted by FEFANA asbl.

2. Formic acid (E 236), Ammonium Formate (E 295), Sodium Formate (E 237) for all animal species. Supplementary information. July 2014. Submitted by FEFANA/HYFAC.

3. Evaluation report of the European Union Reference Laboratory for Feed Additives on the Methods(s) of Analysis for Formic Acid.

4. Evaluation report of the European Union Reference Laboratory for Feed Additives on the Methods(s) of Analysis for Ammonium Formate (E 295) and Sodium Formate (E 237).

5. Comments from Member States received through the ScienceNet.

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Annex A. Executive Summary of the Evaluation Report of the European Union Reference Laboratory for Feed Additives on the Method(s) of Analysis for formic acid[1]

Formic acid is a *feed additive* for which authorisation is sought under the category "technological additive", functional group 1(a) "preservatives" and 1(k) "silage additive", according to Annex I of Regulation (EC) No 1831/2003. In the current application submitted according to Article 4(1) (new use in water) and Article 10(2) of Regulation (EC) No 1831/2003, authorisation for all animal species is requested.

The additive is intended to be marketed as a liquid containing 84.5% *formic acid* and 15.5% water, and to be incorporated into *silages*, complete *feedingstuffs* or *water* (not in *premixtures*) at a maximum *formic acid* concentration of 10, 20 or 5 g/kg, respectively. However, no minimum content was proposed by the Applicant.

For the determination of *formic acid* in *feedingstuffs*, the Applicant suggested a single-laboratory validated method based on ion chromatography with electrical conductivity detection (IC-ECD). The following performance characteristics were reported: - a limit of detection (LOD) and quantification (LOQ) of 100 and 500 mg/kg *feedingstuffs*, respectively; - a recovery rate close to 100 %; and - a relative standard deviation for *repeatability* (RSD_r) of *ca.* 3.5 %. The validation experiments were performed with a set of different feed samples covering a formate content ranging from 3.6 to 10 g/kg. These samples were also analysed by a second independent expert laboratory and all the results were in agreement. Furthermore, the validation report included summary information related to an inter-laboratory comparison organised by VDLUFA in 2006 for the determination of organic acids in *feedingstuffs*, including *formic acid*, to which six laboratories participated. The following performance characteristics were reported, for sample *formic acid* concentrations ranging from 7.2 to 506 g/kg *feedingstuffs*: - RSD_r ranging from 4 to 10%; and - a *reproducibility* relative standard deviation (RSD_R) ranging from 13 to 22%. Based on these acceptable performance characteristics, the CRL recommends for official control purposes the method submitted by the Applicant for the determination of *formic acid* in *feedingstuffs* in the frame of authorisation.

Furthermore, the CRL considers that this method could apply to the determination of *formic acid* in the *feed additive* and *water*, in the frame of this authorisation.

The unambiguous determination of the content of exogenous *formic acid* added to *silages* is not achievable by analysis. The Applicant did not provide any experimental data nor suggested any methods of analysis. Therefore the CRL cannot evaluate nor recommend any method for official control to determine *formic acid* in *silages*.

Based on the several considerations presented, the CRL recommends for official control - in the frame of this authorisation - the single-laboratory validated (and further verified) method submitted by the Applicant for the determination of *formic acid* in *feed additive*, *feedingstuffs* and *water* (not in *silages*).

Further testing or validation is not considered necessary.

[1] The full report is available on the EURL website: http://irmm.jrc.ec.europa.eu/SiteCollectionDocuments/FinRep-FAD-2009-0027.pdf

Annex B. Executive Summary of the Evaluation Report of the European Union Reference Laboratory for Feed Additives on the Method(s) of Analysis for ammonium formate and sodium formate[1,2]

In the current application authorisation is sought under articles 4(1)[3] and 10(2)[4] for *ammonium formate* (E295)[5], *sodium formate* (E237)[6], *calcium formate* (E238)[7]and *potassium diformate* (237a)[8] under the category of "technological additives" functional group 1(a) "preservatives" (for all) and 1(k) "silage additives" (not for *calcium formate* and *potassium diformate*) according to the classification system of Annex I of Regulation (EC) No 1831/2003.

Ammonium formate (E295) is a liquid consisting of a minimum of 35% of ammonium formate and a maximum of 64% of free formic acid, the rest being water. *Sodium formate* (E237) is either a solid with a minimum purity of 98% or a liquid containing a minimum of 15% of sodium formate and a maximum of 75% of free formic acid, the rest being water. Authorisation is sought for the use of these two *feed additives* for all animal species and categories. Both *feed additives* are intended to be used in *premixtures*, *feedingstuffs*, *water* and *silage*. The Applicant proposes a maximum concentration (expressed as formic acid) of 5 g/L in *water*, 10 g/kg in *silage* and 20 g/kg in *feedingstuffs*.

Calcium formate (E238) is a colourless solid with a minimum purity of 98%. Authorisation is sought for the use of the *feed additive* for all animal species and categories. The *feed additive* is intended to be used in *premixtures* and *feedingstuffs*. The Applicant proposes a maximum concentration (expressed as formic acid) of 30 g/kg in *feedingstuffs*.

Finally, *potassium diformate* (237a) is a colourless aqueous solution with a minimum of 50% of potassium diformate in water. Authorisation is sought for the use of the *feed additive* for all animal species and categories. The *feed additive* is intended to be used in *premixtures* and *feedingstuffs*. The Applicant proposes a maximum concentration (expressed as formic acid) of 9, 18 and 12 g/kg in fish for feed, in *feedingstuffs* for weaned piglets and sows & pigs for fattening, respectively.

The following international ring-trial validated standards were submitted by the Applicants and/or identified by the EURL:

- EN ISO 6869 and EN 15510, based on atomic absorption spectrometry (AAS) and inductively coupled plasma atomic emission spectrometry (ICP-AES), respectively, for the determination of total *calcium*, *potassium* and *sodium* in the *feed additives* (*calcium formate* - E238; *potassium diformate* - 237a; *sodium formate* - E237);

- ISO 5664, based on the distillation & titration of ammonia and the Community method (R152/2009, Annex III, C) based on Kjeldahl for the determination of nitrogen and calculation the *ammonium* content in the *feed additive* from the measured nitrogen content (*ammonium formate* - E295);

- EN 15909, based on the EDTA complexometric reaction for the determination of total *calcium* in the *feed additive* (*calcium formate* - E238); and

[1] The full report is available on the EURL website: http://irmm.jrc.ec.europa.eu/SiteCollectionDocuments/FinRep-FormateGroup.pdf
[2] The EURL produced a combined report for the dossier FAD-2010-0188 Potassium diformate for all animal species; dossier FAD-2010-0312 Ammonium formate, sodium formate, calcium formate for all animal species; dossier FAD-2010-0303 Calcium formate for all animal species; and dossier FAD-2009-0027 Formic acid for all animal species.
[3] FAD-2010-0312.
[4] FAD-2010-0312; FAD-2010-0303;FAD-2010-0188.
[5] FAD-2010-0312.
[6] FAD-2010-0312.
[7] FAD-2010-0312; FAD-2010-0303.
[8] FAD-2010-0188.

- EN 15909, based on reverse phase high performance liquid chromatography with UV detection (RP-HPLC-UV) at 214 nm, for the determination of total *formate* in all the *feed additives* of concern (E295, E237, 237a and E238).

The EURL recommends for official control all the above mentioned methods for the determination of *feed additives* of concern. The relevant details about the principles of analysis and the corresponding performance characteristics are provided in the report.

For the determination of *total formate* (originating from *ammonium, sodium, calcium formates* and *potassium diformate*) in *premixtures* and *feedingstuffs* the Applicant[9] proposed a single laboratory validated method based on ion-exclusion high performance liquid chromatography with UV or refractive index detection (HPLC-UV/RI). This method does not distinguish between *formic acid* and its salts. The following performance characteristics for the quantification of *total formate* (expressed as *total formic acid*), were derived from the single-laboratory validation study in *premixtures* and *feedingstuffs*: - a relative standard deviations for *repeatability* (RSDr) ranging from 1.5 to 6.5% for the concentration ranging from 1 to 1000 g/kg; - a *recovery rate* (Rrec) ranging from 89 to 99%; and - a limit of quantification (LOQ) of 90 mg *formic acid*/kg *feedingstuffs*. The HPLC-UV/RI method was further ring trial validated with three to five laboratories and a relative standard deviation for *reproducibility* (RSDR) ranging from 6.6 to 19.3% was determined for *premixtures* and *feedingstuffs* containing from 4.5 to 44 g *formic acid*/kg.

Furthemore, the EURL wishes to recall the conclusions drawn in the frame of the FAD-20090027 "Formic acid" dossier, dated 21/05/2010:

> For the determination of *formic acid* in *feedingstuffs*, the Applicant (FAD-20090027) suggested a single-laboratory validated method based on ion chromatography with electrical conductivity detection (IC-ECD). Approximately 1g of sample is extracted with 80mL of water for 30 minutes and then filled up to 100 mL. After filtration through paper and membrane filters, the solution is injected into the ion chromatograph. External standard calibration is used for the quantification of the formate content. The measured formate content allows the calculation of the formic acid one. The following performance characteristics were reported: - a limit of detection (LOD) and quantification (LOQ) of 100 and 500 mg/kg feedingstuffs, respectively; - a recovery rate (RRec) close to 100 %; and - a repeatability relative standard deviation (RSDr) of ca. 3.5 %. The validation experiments were performed with a set of different feed samples covering a formate content ranging from 3.6 to 10 g/kg. These samples were also analysed by a second independent expert laboratory and all the results were in agreement. Furthermore, the validation report included summary information related to an inter-laboratory comparison organised by VDLUFA in 2006 for the determination of organic acids in *feedingstuffs*, including formic acid, to which six laboratories participated. The following performance characteristics were reported, for sample formic acid concentrations ranging from 7.2 to 506 g/kg *feedingstuffs*: - RSDr ranging from 4 to 10%; and - a reproducibility relative standard deviation (RSDR) ranging from 13 to 22%.

Based on the performance characteristics presented, the EURL recommends for official control the ring trial validated and submitted by Applicant[10] method which is based on ion-exclusion HPLC-UV/RI to determine *total formate* (expressed as *total formic acid*) in *premixtures* and *feedingstuffs* containing *ammonium formate, sodium formate, calcium formate* or *potassium diformate*. In addition, the EURL recommends for official control the IC-ECD method submitted by the Applicant (FAD-20090027) for the determination of *formic acid* (and *total formate*) in *feedingstuffs*. Furthemore, the EURL considers that maximum concentration levels (5g/kg) of *sodium* and *ammonium formates* in *water* could be monitored using the two methods (HPLC-UV/RI or IC-ECD) recommended above.

[9] FAD-2010-0312.
[10] FAD-2010-0312.

None of the Applicants provided analytical method or experimental data for the quantification of *ammonium formate* and *sodium formate* in *silage*. Therefore, the EURL cannot evaluate nor recommend any method for official control to determine *ammonium formate* and *sodium formate* in *silage.*

Further testing or validation of the methods to be performed through the consortium of National Reference Laboratories as specified by Article 10 (Commission Regulation (EC) No 378/2005) is not considered necessary.

The conclusions/recommendations presented in this report for the various *formate* salts are to be combined with those presented in the FAD-2009-0027 for the *formic acid*.