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Safety and efficacy of a feed additive consisting of semi-refined carrageenan for cats and dogs (Gel Systems Ltd.)

EFSA Panel on Additives and Products or Substances used in Animal Feed (FEEDAP), Vasileios Bampidis, Giovanna Azimonti, Maria de Lourdes Bastos, Henrik Christensen, Birgit Dusemund, Mojca Durjava, Maryline Kouba, Marta López-Alonso, Secundino López Puente, Francesca Marcon, Baltasar Mayo, Alena Pechová, Mariana Petkova, Fernando Ramos, Yolanda Sanz, Roberto Edoardo Villa, Ruud Woutersen, Jürgen Gropp, Paola Manini, Joana Firmino, Maria Vittoria Vettori and Matteo Lorenzo Innocenti

Abstract

Following a request from the European Commission, EFSA was asked to deliver a scientific opinion on the safety and efficacy of semi-refined carrageenan as a feed additive for cats and dogs. The EFSA Panel on Additives and Products or Substances used in Animal Feed (FEEDAP) concluded that semi-refined carrageenan was safe for dogs at 6,000 mg/kg final wet feed (with about 20% dry matter). This would correspond to 26,400 mg semi-refined carrageenan/kg complete feed (with 88% dry matter). In the absence of specific data, the maximum concentration of the additive considered safe for cats was 750 mg semi-refined carrageenan/kg final wet feed, corresponding to 3,300 mg semi-refined carrageenan/kg complete feed (with 88% dry matter). In the absence of data, the FEEDAP Panel was not in the position to conclude on the safety of carrageenan for the user. The additive under assessment is intended to be used in dogs and cats only. No environmental risk assessment was considered necessary for such use. The FEEDAP Panel was not in the position to conclude on the efficacy of semi-refined carrageenan as a gelling agent, thickener and stabiliser in feed for cats and dogs at the proposed conditions of use.

Keywords: semi-refined carrageenan, technological additives, gelling agents, stabilisers, thickeners, safety, efficacy

Requestor: European Commission

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Correspondence: feedap@efsa.europa.eu

Panel members: Vasileios Bampidis, Giovanna Azimonti, Maria de Lourdes Bastos, Henrik Christensen, Birgit Dusemund, Mojca Durjava, Maryline Kouba, Marta López-Alonso, Secundino López Puente, Francesca Marcon, Baltasar Mayo, Alena Pechová, Mariana Petkova, Fernando Ramos, Yolanda Sanz, Roberto Edoardo Villa and Ruud Woutersen.

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1. Introduction

1.1. Background and Terms of Reference

Regulation (EC) No 1831/2003[1] establishes the rules governing the Community authorisation of additives for use in animal nutrition. In particular, Article 10(2) of that Regulation also specifies that for existing products within the meaning of Article 10(1), an application shall be submitted in accordance with Article 7, at the latest 1 year before the expiry date of the authorisation given pursuant to Directive 70/524/EEC for additives with a limited authorisation period, and within a maximum of 7 years after the entry into force of this Regulation for additives authorised without a time limit or pursuant to Directive 82/471/EEC.

The European Commission received a request from Gel Systems Ltd[2] for re-evaluation of the product semi-refined carrageenan, when used as a feed additive for cats and dogs (category: technological additives; functional groups: gelling agents, stabilisers, thickeners).

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 10(2) (re-evaluation of an authorised feed additive). EFSA received directly from the applicant the technical dossier in support of this application. The particulars and documents in support of the application were considered valid by EFSA as of 24 April 2014.

According to Article 8 of Regulation (EC) No 1831/2003, 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. EFSA shall deliver an opinion on the safety for the target animals and user, and on the efficacy of the product carrageenan, when used under the proposed conditions of use (see Section 3.1.3).

1.2. Additional information

Semi-refined carrageenan is currently authorised as a feed additive[3] subject to re-evaluation according to Article (10) of Regulation (EC) 1831/2003. Semi-refined carrageenan (with the synonym of processed *Eucheuma* seaweed) is authorised to be used as a food additive in accordance with Annex II to Regulation (EC) No 1333/2008[4] with specific purity criteria defined in Commission Regulation (EU) No 231/2012[5].

Semi-refined carrageenan has not been previously assessed by EFSA as a feed additive. It has been assessed by the Joint FAO/WHO Expert Committee on Food Additives (JECFA, 1972, 1974, 1984, 1987, 1992, 1993, 1995, 1999, 2000, 2001, 2002, 2007a,b, 2008, 2014, 2015) and the Scientific Committee for Food (SCF) (1978, 1983, 1994a,b, 1996, 1998a,b, 2001, 2003a,b). The EFSA Panel on Food Additives and Nutrient Sources added to Food (ANS) delivered an 'opinion on the re-evaluation of carrageenan (E 407) and processed Eucheuma seaweed (E 407a) as food additives' (EFSA ANS Panel et al., 2018).

2. Data and methodologies

2.1. Data

The present assessment is based on data submitted by the applicant in the form of a technical dossier[6] in support of the authorisation request for the use of semi-refined carrageenan as a feed additive.

[1] 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.
[2] Gel Systems Ltd. Elmfield road, 21–23, BR1 1LT, Bromley, Kent, UK.
[3] Commission Directive of 8 July 1985 amending the Annexes to Council Directive 70/524/EEC concerning additives in feedingstuffs, https://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:1985:245:0001:0032:EN:PDF.
[4] Regulation (EC) No 1333/2008 of the European Parliament and of the Council of 16 December 2008 on food additives, https://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2008:354:0016:0033:en:PDF.
[5] Commission Regulation (EU) No 231/2012 of 9 March 2012 laying down specifications for food additives listed in Annexes II and III to Regulation (EC) No 1333/2008 of the European Parliament and of the Council, https://eur-lex.europa.eu/legal-content/EN/TXT/PDF/?uri=CELEX:32012R0231&from=EN.
[6] FEED dossier reference: FAD-2010-0319.

The FEEDAP Panel used the data provided by the applicant together with data from other sources, such as previous risk assessments by EFSA or other expert bodies, peer-reviewed scientific papers, other scientific reports and experts' knowledge, to deliver the present output.

EFSA has verified the European Union Reference Laboratory (EURL) report as it relates to the methods used for the control of semi-refined carrageenan in animal feed.[7]

2.2. Methodologies

The approach followed by the FEEDAP Panel to assess the safety and the efficacy of carrageenan is in line with the principles laid down in Regulation (EC) No 429/2008[8] and the relevant guidance documents: Guidance on studies concerning the safety of use of the additive for users/workers (EFSA FEEDAP Panel, 2012), Guidance on the identity, characterisation and conditions of use of feed additives (EFSA FEEDAP Panel et al., 2017a), Guidance on the assessment of the safety of feed additives for the target species (EFSA FEEDAP Panel et al., 2017b), Guidance on the assessment of the efficacy of feed additives (EFSA FEEDAP Panel et al., 2018) and Guidance on the assessment of the safety of feed additives for the environment (EFSA FEEDAP Panel et al., 2019).

3. Assessment

The product under assessment, semi-refined carrageenan, is a polysaccharide extracted from seaweed of different families belonging to the class of Rhodophyceae (red seaweeds). It is intended to be used as a technological feed additive (functional groups: stabilisers, thickeners, gelling agents) in feedingstuffs for cats and dogs.

3.1. Characterisation

3.1.1. Characterisation of the additive

The additive consists of pure semi-refined carrageenan (synonym of processed *Eucheuma* seaweed), high molecular weight polysaccharides extracted from seaweed of different families belonging to the class of Rhodophyceae (red seaweeds). The main components of semi-refined carrageenan are the polysaccharides kappa- and iota-carrageenan, consisting of the calcium, magnesium, potassium and sodium sulfate esters of galactose and 3,6-anhydrogalactose polysaccharides. Up to 15% of algal cellulose is also present in the product.

Semi-refined carrageenan is manufactured from the main Rhodophyceae species *Eucheuma cottonii* (kappa-carrageenan) and *E. spinosum* (iota-carrageenan). The dry seaweed is treated with strong alkali (potassium hydroxide) at elevated temperature, then washed to remove residual alkali and water-soluble impurities, then dried up to 12% of humidity. The resulting material is then ground to a free-flowing powder.

Ten batches of the additive were analysed with infrared spectroscopy and thin layer chromatography (for the identification of galactose and 3,6-anhydrogalactose),[9] confirming the identity of the additive.[10]

The additive is manufactured to meet the specifications set for the food additive equivalent, processed *Eucheuma* seaweed (E 407a):[11] loss on drying $< 12\%$, viscosity (1.5% solution) at $75°C \geq 5$ mPa·s; methanol ($\leq 0.1\%$); sulfate (as SO_4 on a dry basis) ≥ 15 and $\leq 40\%$; total ash ≥ 15 and $\leq 40\%$; acid insoluble ash (in 10% hydrochloric acid) $\leq 1\%$; low molecular weight carrageenan (molecular weight fraction below 50 kDa) $\leq 5\%$. Semi-refined carrageenan as a feed additive is also

[7] The full report is available on the EURL website: https://ec.europa.eu/jrc/sites/jrcsh/files/finrep-fad-2010-0205%2B0319-carrageenan_amended%20version.pdf.

[8] 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.

[9] As described in the Joint FAO/WHO Expert Committee on Food Additives (JECFA) monograph on Processed Eucheuma seaweed which was prepared at the 68th JECFA (2007).

[10] Technical dossier/Supplementary Information September 2015/Response to EFSA September 7 2015 complete.

[11] Commission Regulation (EU) No 231/2012 of 9 March 2012 laying down specifications for food additives listed in Annexes II and III to Regulation (EC) No 1333/2008 of the European Parliament and of the Council.

specified to contain ≤ 15% acid-insoluble matter (in 1% v/v sulfuric acid). The analysis of five batches of the additive[12,13] showed compliance with the specifications.

Ten additional batches of semi-refined carrageenan (five mainly composed of kappa-carrageenan and five mainly composed on iota-carrageenan) were analysed for viscosity. All the batches showed compliance with the specifications (range 45 to 114 mPa•s and 25 to 54 mPa•s for kappa- and iota-carrageenan, respectively).[14] The same five batches of kappa-carrageenan were also analysed (four replicated analysis) for gel strength (solution of 1% semi-refined carrageenan in a 0.2% potassium chloride solution). The results showed a gel strength in the range of 216 to 479 g.[14]

The analysis of five batches of the additive showed average concentrations of lead of 1.2 mg/kg (range 1–2 mg/kg), cadmium of 0.8 mg/kg (range 0.6–1 mg/kg) and arsenic of 2.38 mg/kg (range 1.8–2.9 mg/kg); mercury concentration was below the limit of quantification, (LOQ < 0.30 mg/kg).[15] *E. coli* (in 10 g) and *Salmonella* spp. (in 25 g) were not detected in any of the batches; total plate count varied between 1.0×10^1 to 2.8×10^4 CFU/g; *C. perfrigens*, filamentous fungi and yeasts were $< 1.0 \times 10^1$ CFU/g in all the batches.[16] Five selected pesticides, analysed in one batch[17] of the additive, were below the respective limits of detection (LODs).[18] The analysed impurities do not raise safety concerns.

No analytical data were provided on possible presence of dioxins and dioxins like PCBs, botanical impurities and mycotoxins. The analysed impurities do not raise safety concerns.

The sieve analysis of five batches of the additive showed that 100% of the particles has dimension < 600 μm, and that 4% to 9% of the particles has dimensions < 53 μm.[19] The particles size analysis of three batches of the additive, done with laser diffraction, showed that 10% of the particles are smaller than 27–55 μm.[20] No information on the presence of particles smaller than 1 μm, including nanoparticles, was provided. Three batches of the additive (two mainly composed of kappa-carrageenan and one mainly composed on iota- carrageenan) were analysed (triplicate analysis) for dusting potential (Stauber-Heubach method).[14] The batch of iota-carrageenan showed a dusting potential of 12.8 g/m^3; the two batches of kappa-carrageenan showed dusting potentials of 4.1 and 18.8 g/m^3, respectively.

3.1.2. Stability and homogeneity

Three batches of the additive (mainly composed of kappa-carrageenan) were stored for 3 months in closed packages at 25°C and 60% RH, or at 40°C and 75% RH.[21] The samples were analysed every 6 weeks for moisture content; solutions of 1.5% semi-refined carrageenan in water or 1.2% semi-refined carrageenan in 0.3% potassium chloride were prepared and analysed at the same timepoints for viscosity. The gels formed with these solutions were then analysed for gel strength and extensibility. No major differences were observed during the trial between the different solutions and respective gels at any of the storing conditions.

For technological additives, stability in feedingstuffs can be demonstrated by the persistence of the effect; no demonstration of homogenous distribution is considered necessary if the efficacy of the additive is demonstrated. The applicant has provided three studies aimed to show the effects of semi-refined carrageenan on the gel strength of feedingstuffs, described in Section 3.3.

3.1.3. Conditions of use

The additive is intended to be used as a technological additive (functional groups: gelling agents, stabilisers and thickeners) in feedingstuffs for cats and dogs, with no minimum or maximum content. The applicant proposed typical use levels of 1,000–20,000 mg/kg of semi-refined carrageenan in the final wet feed.

[12] Technical dossier/Section II/Annexes Sect. II/Annex II_1b Cof A_.
[13] Technical dossier/Section II/Annex_II_1c MW Report.
[14] Technical dossier/Supplementary Information September 2015/Response to EFSA September 72,015 complete.
[15] Technical dossier/Section II/Annex_II_1d HM CofA.
[16] Technical dossier/Section II/Annex_II_1e Micro CofA.
[17] Technical dossier/Section II/Annex_II_1f Pesticides.
[18] Limits of detection (LODs): glyphosate, 0.1 mg/kg; chlorpyriphos-methyl, 1.0 mg/kg; malathion, < 1.0 mg/kg; methamidophos, < 1.0 mg/kg; pirimiphos-methyl, 1.0 mg/kg.
[19] Technical dossier/Section II.
[20] Technical dossier/Section II/Annex_II_1g Particle size.
[21] Technical dossier/Section II/Annex_II_3_Stability.

3.2. Safety

To support the safety for the target species, the applicant submitted information from the literature, as well as original reports of toxicological studies. All these studies were reviewed in previous assessments by other assessment bodies. In particular, the same toxicological dataset submitted has been previously assessed by the EFSA ANS Panel in 2018 in an opinion on the re-evaluation of carrageenan (E 407) and processed *Eucheuma* seaweed (E 407a) as food additives (EFSA ANS Panel et al., 2018). The ANS Panel concluded that '… the absorption, distribution, metabolism and excretion (ADME) database was sufficient to conclude that carrageenan was not absorbed intact; in a sub-chronic toxicity study performed with carrageenan almost complying with the EU specification for E 407 in rats, the no-observed-adverse-effect level (NOAEL) was 3,400–3,900 mg/kg bw per day, the highest dose tested; no adverse effects have been detected in chronic toxicity studies with carrageenan in rats up to 7,500 mg/kg bw per day, the highest dose tested; there was no concern with respect to the carcinogenicity of carrageenan; carrageenan and processed Eucheuma seaweed did not raise a concern with respect to genotoxicity; the NOAELs of sodium and calcium carrageenan for prenatal developmental dietary toxicity studies were the highest doses tested; the safety of processed Eucheuma seaweed was sufficiently covered by the toxicological evaluation of carrageenan; data were adequate for a refined exposure assessment for 41 out of 79 food categories. However, the ANS Panel noted uncertainties as regards the chemistry, the exposure assessment and biological and toxicological data. Overall, taking into account the lack of adequate data to address these uncertainties, the Panel concluded that the existing group acceptable daily intake (ADI) for carrageenan (E 407) and processed Eucheuma seaweed (E 407a) of 75 mg/kg bw per day should be considered temporary, while the database should be improved within 5 years after publication of this opinion'.

The FEEDAP Panel, having reviewed the relevant toxicological studies done with carrageenan and semi-refined carrageenan, in agreement with the assessment of the EFSA ANS Panel, concluded that semi-refined carrageenan does not raise a concern for genotoxicity, carcinogenicity and reproductive toxicity.

3.2.1. Safety for the target species

The maximum safe concentration of the additive in complete feed for cats and dogs could be derived using the results of the toxicological studies (EFSA FEEDAP Panel et al., 2017b). Among the studies evaluated in the assessment, the FEEDAP Panel considered that the results of the chronic toxicity study in rats are the most appropriate to identify a NOAEL (7,500 mg/kg bw and day). An uncertainty factor of 100 was used to cover the intra- and interspecies variation. The results are reported in Table 1.

Table 1: Maximum safe concentration in feed of semi-refined carrageenan for cats and dogs

Animal category	Default values		Maximum safe concentration in feed (mg/kg feed)[1]
	Body weight (kg)	Feed intake (g DM/day)	
Dog	15	250	4,000
Cat	3	60	3,300

[1]Complete feed dry matter (DM) = 88%.

The maximum safe concentrations in complete feedingstuffs for cats and dogs (3,300 and 4,000 mg/kg complete feed, respectively), would correspond to approximately 750 and 900 mg/kg final wet feed (with about 20% dry matter).

To support the safety at the proposed conditions of use, the applicant submitted a tolerance trial with dogs.

3.2.1.1. Tolerance study in dogs

The blinded study[22] was started with a total of 30 Beagle dogs (equal numbers of male and females, between 1 and 11 years old, body weight 10.9–19.0 kg) after a 7-day acclimation phase, in which the dogs were accustomed to a commercial wet diet. The dogs were randomly allocated to one of the 3 dietary treatments ensuring each group was balanced for sex and weight resulting in 5 dogs per sex and group. After a pre-period of 7 days, two dogs of each group with the lowest feed intake were removed regardless of sex, and the 28-day tolerance study started.

From study-day −7 until study-day 28 included, the dogs were individually housed for a period of up to 4 h each day to facilitate individual feeding, diet consumption assessment, faecal consistency scoring and clinical assessment. Animals were then returned to group housing, where they were housed with animals allocated to the same study group.

The experimental diets, nutritionally balanced and mainly consisting of water, lungs, chicken MDM,[23] liver and a flavour source, containing by analysis 79% moisture, about 9% CP and 3.3 MJ ME/kg (calculated), all supplemented with gelling agents at a concentration of 60,000 mg/kg final wet feed. The control diet was supplemented with an authorised gelling agent, the use level diet (1×) was supplemented with 6,000 mg/kg semi-refined carrageenan and 54,000 mg of the other authorised gelling, and the overdose diet was supplemented with 60,000 mg/kg semi-refined carrageenan (10×). These inclusion levels of semi-refined carrageenan would correspond to approximately 26,400 and 264,000 mg/kg complete feed (with 88% of dry matter). The diets were fed according to the dog's energy requirement, based on the calculated energy values for the first 2 weeks; the amount was increased for weeks 3 and 4 by about 10%. The FEEDAP Panel considers this design acceptable to test any adverse effects of the additive under assessment.

Clinical assessments were performed daily, approximately 4 h post administration of the test diets, and included restlessness, lethargy, vomitus and faeces. Body weight (prior to feeding) and body condition score were determined in weekly intervals. Feed intake was measured daily. Blood samples for routine haematology[24] and biochemistry[25] analyses were collected from all animals at study-day −7, at study start and study end.

The experimental unit was the animal. Body weight, body condition score and feed intake as well as haematology and biochemistry endpoints were analysed using a mixed model analysis of variance for repeated measure design. The data per day were assessed by ANOVA (two-way ANOVA, including sex) for haematology and blood biochemistry.

No abnormal general health observations were recorded for any animal at any time point for restlessness or lethargy; vomitus in one dog of each group occurred in the pre-period.

Weekly body weight was not different among the groups (Table 2). The repeated measure analysis identified a highly significant effect of time, with a distinct decrease of body weight from day −7 to study start and to day 14 within a group. The decrease on body weight is seen as a consequence of an insufficient energy supply; dietary energy was calculated by table values not considering the effect of gelatinising binders (which are hardly digestible) on the digestibility of other nutrients. The increase in the amount of feed offered stopped this decrease for the control group and the use level group, but not for the 10× group. The continued loss of body weight in the second 14 study-days of the 10× overdose group (less accentuated than in the first 14-day period) is considered as a nutrient binding/reduced digestibility effect of the additive, not included in the calculation of dietary energy content, and thus resulting in a lower allocation of energy than required. Consequently, the effect on body weight in the overdose group is not considered adverse. The weekly and total feed intake did not differ among the groups, while the effect of time was significant; however, this was to be expected, considering the increased amount of feed given in the second half of the tolerance study.

[22] Animals were housed and maintained according to the requirements of the European Directive 2010/63/EU (EC, 2010). The animal facility in which the study was conducted is authorised for such studies by the Health Products Regulatory Authority of Ireland.

[23] Mechanically deboned chicken.

[24] Red blood cells (RBC), haemoglobin, packed cell volume (PCV), haematocrit (HCT), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), platelets, white blood cells (WBC), neutrophils, lymphocytes, monocytes, eosinophils, and reticulocyte count.

[25] Total protein (TP), albumin, globulin, sodium, potassium, chloride, calcium, magnesium, phosphate, glucose, cholesterol, bilirubin, urea, creatinine, alkaline phosphatase (ALP), alanine transaminase (ALT), creatinine kinase (CK) lactate dehydrogenase (LDH) and gamma-glutamyl transferase (GGT).

Table 2: Body weight of dogs fed different levels of semi-refined carrageenan at the expense of locust bean seed flour in a tolerance study of 28 days duration after a pre-period of 7 days

Semi-refined carrageenan inclusion level (mg/kg wet food)	Body weight (kg)			
	Day −7	Day 1	Day 14	Day 27
0	14.1	13.5	13.0	13.1
6,000	14.3	13.7	13.3	13.3
60,000	14.1	13.5	13.0	12.8

Body condition score, with scores from 1 (no discernible body fat, loss of muscle mass) to 5 (ribs palpable without excess of fat covering), did not show differences among the groups at all days of the assessment. However, the effect of time was significant, with a decrease of the score from values between 4.75 and 5.00 at the begin of the tolerance study, and between 4.13 and 4.25 at study end; this effect was also to be expected, considering the loss of weight in the first part of the tolerance study.

Abnormal faeces consistency, indicated by faecal scores of 4 (poorly formed, viscous), 5 (diarrhoea) or 6 (watery diarrhoea), was seen on several occasions in 6 dogs each of the control group and the 10× group, and in 7 dogs of the use level group.

At study-day −7, no significant group differences were seen for the haematological endpoints (except for platelets with higher value in the control group) and at the end of the tolerance study, haematology results displayed no significant treatment or interaction (treatment × sex) effects.

Regarding blood biochemistry values at study-day −7, there were no significant differences among treatment groups except for total protein, which was lower for the dogs of the control group compared to those in the use level and the 10× groups. Most endpoints for clinical routine biochemistry at study end did not reflect significant treatment effects. Globulins, phosphate, urea and gamma-glutamyl transferase were significantly lower ($p < 0.05$) in female dogs (31.2 g/L, 0.57 mmol/L, 4.1 mmol/L, 3.7 U/L, respectively) when compared to the male dogs (35.7 g/L, 0.73 mmol/L, 4.5 mmol/L, 6.1 U/L, respectively), remaining within the reference range (except phosphate).

The results for the endpoints showing differences at the end of the study are reported in Table 3.

Table 3: Selected blood biochemistry parameters of dogs fed different levels of semi-refined carrageenan at the expense of locust bean seed flour at study end (day 28)

Semi-refined carrageenan inclusion level (mg/kg wet food)	Blood biochemistry parameters				
	Potassium (mmol/L)	Sodium (mmol/L)	Magnesium (mmol/L)	Alkaline phosphatase (U/L)	Alanine transaminase (U/L)
0	4.5[a]	145.7[a]	0.76[a]	84.1[a]	198.0[a]
6,000	4.5[a]	144.5[b]	0.68[b]	47.0[b]	170.9[a]
60,000	4.3[b]	145.6[a]	0.71[ab]	51.3[b]	73.4[b]

a,b: Mean values within a column with a different superscript are significantly different ($p < 0.05$).

All values for sodium, potassium, magnesium and alkaline phosphatase (ALP) remained within the reference range.[26] Considering that the differences for sodium, potassium and magnesium blood levels were very small, that those for sodium and magnesium did not appear as dose dependent, and all values were within the reference range, these findings were not considered biologically relevant.

Dogs fed diets containing 6,000 (1×) or 60,000 mg (10×) semi-refined carrageenan/kg wet feed displayed significantly lower ALP activity than the control group. At day −7, ALP activities were already lower in dogs fed the supplemented diets relative to the controls (63.9 and 62.3 vs. 88.0 U/L), and the difference reached significance in the 10× group by start of the tolerance study (81.3 vs. 54.5 U/L). It is very likely that the change in ALP activity was not attributable to the level of semi-refined carrageenan in the diet.

[26] Combined reference intervals communicated by MSD Manual Synlab UK, UCD Veterinary Laboratory and the institution where the study has been conducted.

At study end, alanine transaminase (ALT) activities of the 10× overdose group were significantly lower than in the use level and control groups. However, only the values of the overdose group were within the reference range (10.0–109.0 U/L). At study-day −7, ALT activities were relatively consistent among treatment groups (38.8, 40.0 and 31.3 U/L), but were numerically elevated in the control group (79.1 U/L vs. 53.9 and 42.4 U/L for the use level and the 10× group, respectively) at the start of the tolerance study. The higher values of the control group and of the use level group could rather be attributed to the other authorised gelling agent included in the diets.

3.2.1.2. Conclusions on safety for the target species

The results of a 28-day tolerance study in dogs indicate that 6,000 mg semi-refined carrageenan/kg final wet feed is safe in a diet with about 20% dry matter. This corresponds to 26,400 mg semi-refined carrageenan/kg complete feed (with 88% dry matter). The margin of safety is 10.

In the absence of a specific tolerance study, the maximum concentration of the additive considered safe for cats is 750 mg semi-refined carrageenan/kg final wet feed (with about 20% dry matter), corresponding to 3,300 mg semi-refined carrageenan/kg complete feed (with 88% dry matter).

3.2.2. Safety for the user

No specific information was provided by the applicant. In the absence of data, the FEEDAP Panel is not in the position to conclude on the safety of semi-refined carrageenan for the user.

3.2.3. Safety for the environment

The additive under assessment is intended to be used in cats and dogs only. No environmental risk assessment is necessary for such use (EFSA FEEDAP Panel et al., 2019).

3.3. Efficacy

The applicant has provided three studies to support the stability/efficacy of semi-refined carrageenan as a gelling agent, stabiliser and thickener.[14] In the three studies, a blend of gelling agents was used.

In the first study, [redacted]

In the second study, [redacted]

In the third study, [redacted]

The three studies showed several deficiencies, in particular: (i) the pet foods used were not identified, nor the composition and the DM content reported; (ii) no statistical analysis of the results was performed; (iii) the additive was used in combination with other gelling agents; (iv) no control group without the additive was included in the design of the studies. Due to these shortcomings, the FEEDAP Panel is not in the position to conclude on the efficacy of semi-refined carrageenan as a gelling agent, thickener and stabiliser in feed for cats and dogs at the proposed conditions of use.

4. Conclusions

Semi-refined carrageenan is safe for dogs at 6,000 mg/kg final wet feed (with about 20% dry matter). This corresponds to 26,400 mg semi-refined carrageenan/kg complete feed (with 88% dry matter). In the absence of specific data, the maximum concentration of the additive considered safe for cats is 750 mg semi-refined carrageenan/kg final wet feed (with about 20% dry matter), corresponding to 3,300 mg semi-refined carrageenan/kg complete feed (with 88% dry matter).

In the absence of data, the FEEDAP Panel is not in the position to conclude on the safety of carrageenan for the user.

The additive under assessment is intended to be used in dogs and cats only. No environmental risk assessment is necessary for such use.

The FEEDAP Panel is not in the position to conclude on the efficacy of semi-refined carrageenan as a gelling agent, thickener and stabiliser in feed for cats and dogs at the proposed conditions of use.

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Abbreviations

ADI	average daily intake
ADME	absorption, distribution, metabolism and excretion

ANS	EFSA Scientific Panel on Additives and Nutrient Sources added to Food
bw	body weight
CAS	Chemical Abstracts Service
CFU	colony forming unit
CV	coefficient of variation
DM	dry matter
EINECS	European Inventory of Existing Chemical Substances
EURL	European Union Reference Laboratory
FAO	Food Agricultural Organization
JECFA	The Joint FAO/WHO Expert Committee on Food Additives
LOD	limit of detection
LOQ	limit of quantification
NOAEL	no observed adverse effect level
SD	standard deviation
WHO	World Health Organization