FOOD STANDARDS Australia New Zealand
Te Mana Kounga Kai – Ahitereiria me Aotearoa

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Supporting document 1

Risk and technical Assessment Report – A1126

Pectins & Carrageenan as Processing Aids in Wine (Fining Agent)

Executive summary

FSANZ has assessed the evidence on the safety of pectins and carrageenan as processing aids in wine. The data provided with the Application, together with information from other sources, are considered adequate for hazard assessment.

Both pectins and carrageenan have a long history of safe use in the human food supply, and are approved as food additives at GMP, in Schedule 16 – Types of substances that may be used as food additives in the *Australia New Zealand Food Standards Code* (Code).

The evidence presented to support the proposed use provides adequate assurance that pectins and carrageenan are technologically effective as fining agents in wine manufacture and provide alternatives to the use of bentonite for this purpose. The residual levels in wine are very low and do not perform any technological purpose in the final product. There are identity and purity specifications for pectins and carrageenan in the primary reference sources listed in Schedule 3 – Identity and purity.

Pectins are resistant to digestion in the small intestine but are broken down by intestinal flora, with no adverse effects of metabolites identified. Carrageenan is not broken down in the gastrointestinal tract. Neither pectins nor carrageenan are genotoxic.

A review of recent animal and human studies by JECFA in 2015 did not identify any adverse effects relevant to human health risk assessment for either pectins or carrageenan.

Allergic reactions to ingested pectins appear to be extremely rare and may represent cross-reaction with allergens of cashew nuts and/or pistachio nuts. There is no robust evidence of allergic reactions to ingested carrageenan.

Based on the reviewed toxicological data, it is concluded that in the absence of any identifiable hazard, an Acceptable Daily Intake (ADI) 'not specified' remains appropriate for both pectins and carrageenan.

In conclusion, pectins and carrageenan achieve the technological function in the quantity and form proposed to be used as a food processing aid, and there are no public health and safety concerns from the use of pectins and carrageenan as processing aids in the manufacture of wine in Australia.

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1 Background

1.1 Chemistry

1.1.1 Pectins

Pectins are extracted from plant material, most commonly the peels of apples or citrus fruits.

Pectins, or pectic oligosaccharides, principally consist of the partial methyl esters of polygalacturonic acid in an alpha (1-4) chain and their sodium, potassium, calcium and ammonium salts. Pectins may contain acetate or other ester groups. Commercial pectins typically have a molecular weight in the range 60 to 130,000 Da.

Pectins have been assigned the CAS number 9000-69-5 and the EINEC number 232-533-0. In the International Numbering System (INS) pectin is 440. In Europe, pectins are classified as either 440(i) for non-amidated pectins and 440(ii) for amidated pectins.

When added to water, powdered pectins form a gel.

1.1.2 Carrageenan

Carrageenan is manufactured from red seaweeds of the class *Rhodophyceae*.

Carrageenans are highly sulphated galactans. Carrageenans are classed into three major families based on the position of sulphate groups on their 1,3- and 1,4-linked disaccharide galactose residues. Kappa (κ) carrageenan has one sulphate per disaccharide, iota (ί) carrageenan has two sulphates per disaccharide, and lambda (λ) carrageenan has three sulphates per disaccharide. Commercial carrageenan may be the sodium, potassium, calcium or ammonium salt. Carrageenans have molecular weights in the range 200,000 – 800,000 Da.

Carrageenans have been assigned the CAS number 11114-20-8, the EINEC number 232-534-2, and the INS number E407.

Carrageenans vary in their gelling properties, depending on the number and position of the sulphate groups. The strength of the gel is inversely related to the number of sulphate groups, and lambda carrageenans are non-gelling.

All carrageenans are soluble in hot water. Lambda carrageenans are soluble in cold water, but kappa and iota carrageenans are soluble in cold water only as their sodium salts.

2 Risk assessment questions

The key risk assessment questions to be addressed are as follows:

1.	Is the proposed technological purpose clearly stated, and do pectins and carrageenan achieve the technological function in the quantity and form proposed to be used as a food processing aid?
2.	Are there any public health and safety concerns that may arise from the use of pectins and carrageenan as processing aids in the manufacture of wine in Australia?

2.1 Scope

The Application seeks an amendment to the Code to permit pectin and carrageenans to be used as processing aids in wine produced in Australia via amendment of Standard 4.5.1 Wine Production Requirements (Australia only).

Pectins and carrageenan are already permitted processing aids as defined in Standard 1.3.3 – Processing aids, by virtue of being food additives permitted at GMP in Schedule 16 – Types of substances that may be used as food additives . Therefore, the current assessment is generally limited to their specific technical function as fining agents in wine manufacture and a review of recent data on the toxicology of pectin and carrageenans.

Pectins as food additives have been evaluated by the Joint FAO/WHO Expert Committee on Food Additives (JECFA) at its 13th, 17th, 18th, 19th and 25th meetings. A group acceptable daily intake (ADI) of 'not specified' was established for pectin and amidated pectin in 1981 at the 25th meeting. An addendum was prepared as a result of the 79th JECFA meeting, and published in 2015 (WHO 2015).

Carrageenan was evaluated by JECFA at its 13th, 17th, 28th, 51st, 57th and 68th meetings. An ADI of 'not specified' was established at the 28th meeting. At its 44th meeting, JECFA extended this ADI to include processed *Eucheuma* seaweed at the 51st meeting, but made the ADI temporary for carrageenan and processed *Eucheuma* seaweed, pending clarification of the significance of promotion of colon cancer in rats. The group ADI for carrageenan and processed *Eucheuma* seaweed was finalised at the 57th meeting.

At that meeting, JECFA concluded that continuous feeding of high doses of carrageenan caused a generalised proliferative response in the colonic mucosa of male rats, a response that might play a role in the observed promotion by high doses of carrageenan of the tumourigenicity of known colon carcinogens. However, JECFA noted that the enhancement of colon carcinogenesis in rats was seen only at a dietary concentration far exceeding likely human intake, and that carrageenan at 50 g/kg of diet did not promote tumours in rat colon in a classical initiation–promotion study. JECFA therefore considered that intake of carrageenan and processed *Eucheuma* seaweed was not of concern and allocated a group ADI of 'not specified' for carrageenan and processed *Eucheuma* seaweed. An addendum was prepared as a result of the 79th JECFA meeting, and published in 2015 (WHO 2015).

The addenda from the 79th meeting relate to the use of pectins or carrageenan in infant formulas, but are nevertheless relevant to this Application because they include new scientific evidence for the general safety of pectins and carrageenan.

2.2 Evaluation of submitted data

FSANZ has assessed the submitted evidence on the use of pectins and carrageenan as wine fining agents plus the safety of pectins and carrageenan including information on history of safe use. Together with information from the JECFA addenda published in 2015, and some other published papers and reviews, the submitted data are considered suitable for hazard assessment for pectin and carrageenan.

3 Food technology assessment

3.1 Specifications

Section 15 of Standard 1.1.1 – Structure of the Code and general provisions requires that, with some exceptions, all substances used as processing aids must comply with any relevant specification set out in Schedule 3. Schedule 3 states that the primary sources are specific editions of FAO JECFA Monographs, the United States Food Chemicals Codex, or Commission Regulation (EU) No 231/2012.

These sources contain specifications for pectins and carrageenan.

3.2 Technological function

Pectins and carrageenan are negatively charged polysaccharides. They are commonly used as food additives with the functions of emulsifier, gelling agent, glazing agent, stabiliser or thickener.

However, their ability to form complexes with positively charged molecules, including proteins, means that they can also be used as processing aids to assist in the removal of the positively charged components through filtration or settling. As noted above, pectins and carrageenan are already permitted processing aids as defined in Standard 1.3.3, by virtue of being food additives permitted at GMP in Schedule 16, including, for example, in the fining of beer.

The Application is for the specific inclusion of pectins and carrageenan as processing aids in Standard 4.5.1, the scope of which is the production of wine in Australia, and so this is the only use considered with respect to technological function.

Laboratory and industrial-scale trials were conducted on the use of pectins and/or carrageenan as fining agents during wine manufacture (Marangon et al. 2012). The trials showed that pectins and/or carrageenan were effective in removing protein and protein haze and that the level of bentonite required as a fining agent was significantly reduced. The process of settling resulted in the levels of polysaccharides in the final wine being the same as when pectins and carrageenans were not used, with the authors concluding “that there was little chance of having residual polysaccharides of non-grape origin in the finished wines”.

3.3 Food technology conclusion

The evidence presented to support the proposed use provides adequate assurance that pectins and carrageenan are technologically effective as fining agents in wine manufacture and provide alternatives to the use of bentonite for this purpose. The residual levels in wine are very low and do not perform any technological purpose in the final product. There are identity and purity specifications for pectins and carrageenan in the primary reference sources listed in Schedule 3 – Identity and purity.

4 Hazard assessment

4.1 History of human exposure and consumption

4.1.1 Pectins

Pectins have a longstanding history of safe use in food, for the purposes of gelling, thickening and stabilising.

Pectins also have a long history of use in pharmaceuticals (May 1990).

4.1.2 Carrageenan

Carrageenan also has a long history of safe use in food, for the purposes of thickening, gelling, stabilizing and glazing.

Carrageenan gel matrices have industrial and pharmaceutical applications for the immobilization of bacteria and yeasts (Necas and Bartosikova 2013).

4.2 Metabolism

4.2.1 Pectin

Pectins are resistant to digestion in the small intestine, and are metabolized by microflora in the large intestine. *In vitro*, pectins are metabolized to oligogalacturonic acid, which is further metabolized to acetate, propionate and butyrate (WHO 2015). Methanol is produced as a result of the large intestinal fermentation of pectins, but human consumption of large quantities (up to 50 g/day) of pectins is not associated with any adverse effects that could be attributed to methanol (Siragusa et al. 1988).

4.2.2 Carrageenan

Studies in a variety of species of experimental animals, including rats, guinea pigs and primates, have shown that there is negligible absorption of food grade carrageenan from the gastrointestinal tract (Weiner, 2014; WHO 2015).

4.3 Genotoxicity studies

4.3.1 Pectins

On the basis of recent (2010) genotoxicity studies conducted under GLP and in accordance with OECD guidelines, JECFA (WHO 2015) concluded pectins are not genotoxic. The studies included the bacterial reverse mutation assay (Ames test) ± S9 fraction; mouse lymphoma assay, chromosome aberration assays (pulse and continuous) in Chinese hamster ovary cells, and a micronucleus test in rats. The dose in the rat micronucleus test was 7 g/kg bw/day.

4.3.2 Carrageenan

Carrageenan has been extensively evaluated for genotoxicity using a range of *in vitro* and *in vivo* assays, including the bacterial reverse mutation assay ± S9 fraction, sister chromatid exchange assays, cytogenetic assays, rec-assay in *Bacillus subtilis*, mouse micronucleus assay, host-mediated assays and dominant lethal assays in rats. Results of these various assays have been negative (Cohen and Ito 2002).

4.4 Studies in experimental animals

4.4.1 Pectins

No new data on acute toxicity, long-term toxicity or carcinogenicity of pectins were identified at the 79th JECFA meeting.

A one-generation GLP dietary study in Wistar rats, which included a 13-week subchronic study of the F1 offspring, was reviewed. It was concluded that no effects relevant to humans were found at doses up to and including the highest dose tested, which was 7.1 g/kg bodyweight/day. In two 13-week dietary studies of pectin-derived acidic oligosaccharides (pAOS) in rats, treatment-related diffuse hyperplasia of the urinary bladder epithelium was observed, in conjunction with increased urinary pH and sodium concentration. This is a well-recognized species-specific effect of increased urinary pH and sodium concentration in rats, which is not relevant to human risk assessment. The NOAEL of pAOS in these studies was approximately 7 g/kg bw/day, the highest dose tested (WHO 2015).

JECFA also reviewed two studies of pectin conducted in pigs. In a study of the effects of dietary fibre on intestinal morphology, conducted over nine days in weanling pigs, inclusion of 7.1% pectin in the diet was associated with decreased feed intake and bodyweight gain, and with shorter crypts and increased crypt density in the small intestine (Hedemann et al., 2006, as reviewed by WHO 2015). A GLP-compliant study of pectin in milk replacer, administered in milk to neonatal pigs for three weeks, resulted in decreased bodyweight gain, decreased feed efficiency and, in males, decreased milk replacer consumption at the highest dose of pectin, 10 g/L milk replacer, approximately equal to 3013 mg/kg bw/day for males. These effects were correlated with changes in intestinal microbiota. The NOAEL was identified as the middle dose, 3 g/L, approximately equal to 847 mg/kg bw/day (MPI, 2013, as reviewed by WHO 2015).

4.4.2 Carrageenan

New studies in animals identified at the 79th meeting of JECFA were limited to studies in neonatal and juvenile pigs, and are therefore not directly relevant to the current application. However, no treatment-related effects were found in any parameter in unweaned piglets at dietary carrageenan concentrations up to 2250 ppm, equivalent to approximately 430 mg/kg bw/day (WHO 2015).

4.5 Studies in humans

4.5.1 Pectins

New studies of pectins in humans reviewed by JECFA at the 79th meeting were limited to studies in infants and therefore not directly relevant to the current application. No adverse effects were observed as a result of the inclusion of pectins in infant formula at approximately 240 mg/kg bw/day for up to six weeks (WHO 2015).

4.5.2 Carrageenan

At the seventy-ninth meeting of JECFA, only two brief reports of studies of carrageenans in infants were noted. No adverse effects were noted as a result of consumption of carrageenan in formula at 300 mg/L for 6 months, or 1000 mg/L for 112 days (WHO 2015).

4.6 Potential for allergenicity

Allergic reactions to ingested pectins appear to be extremely rare and may represent cross-reaction with allergens of cashew nuts and/or pistachio nuts (Ferdman et al. 2006). There is no robust evidence of allergic reactions to ingested carrageenan (WHO 2008).

4.6.1 Discussion

Pectins and carrageenan have a long history of safe use in the human food supply.

Pectins are resistant to digestion in the small intestine but are broken down by large intestinal flora, with no adverse effects of metabolites identified. Carrageenan is not broken down in the gastrointestinal tract.

There is no evidence of genotoxicity of pectins or carrageenan.

A review of recent animal and human studies by JECFA in 2015 did not identify any adverse effects relevant to human health risk assessment for either pectins or carrageenan.

5 Conclusions

Based on the reviewed toxicological data, it is concluded that in the absence of any identifiable hazard, an Acceptable Daily Intake (ADI) 'not specified' remains appropriate for both pectins and carrageenan.

Pectins and carrageenan achieve the technological function in the quantity and form proposed to be used as a food processing aid, and there are no public health and safety concerns from the use of pectins and carrageenan as processing aids in the manufacture of wine in Australia

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