

Pine nuts and pine mouth

Emerging issues paper

June 2012

Prepared by the NSW Food Authority on
behalf of the Coordinated Food Survey Plan

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Executive summary

In the past decade, cases of a taste disturbance referred to as 'pine mouth' or 'pine nut syndrome' have been attributed to the consumption of pine nuts. Not all people who consume pine nuts become afflicted with the taste disturbance, but those that do report a bitter or metallic taste lasting from a few days up to two weeks. Cases of pine mouth are not common, however since 2009 there appears to have been a spike in numbers internationally with several hundred complaints lodged with agencies across countries such as France, UK and the USA. Social media blogs tend to indicate the true extent of the issue may be even more widespread.

Mixing of different pine nuts species for retail sale is common practice, which makes identification of any implicated pine nuts difficult. However, several researchers have indicated that a particular pine nut species *Pinus armandii* exported from the Shaanxi and Shanxi regions of China may be responsible for causing the symptoms, as this species of pine nut was not traditionally considered edible.

The actual mechanism of pine mouth has not been determined, however the Codex Alimentarius Commission have moved to exclude *P. armandii* as well as *P. massoniana* from the list of edible tree nuts. In response to the increased incidence of pine mouth cases, the Chinese authorities have implemented measures to accredit exporters of pine nuts and ensure *P. armandii* are no longer exported to the European Union, one of the major consumers of Chinese pine nuts.

Due to the small number of cases reported in Australia, it is recommended that Australian authorities evaluate the ongoing effectiveness of the measures implemented by the Chinese authorities. Any future cases of pine mouth that are reported should be recorded and the source or species of any implicated pine nuts established. If reported cases of pine mouth significantly increases in Australia, consideration could be given to introducing regulatory measures to list implicated pine nut species as prohibited from being offered for sale as food.

1. Introduction

1.1 First reported case of pine mouth

The first documented case of pine mouth in the scientific literature dates back to 2001 when Mostin (2001) published an article in the *European Journal of Emergency Medicine*. He noted that a colleague at the Poisons Centre in Brussels had experienced several episodes of taste disturbance a few days after consuming pine nuts. The phenomenon was described as a bitter, metallic taste disturbance usually lasting a few days. Since that time, this taste disturbance has become referred to as 'pine mouth' or 'pine nut syndrome', however medically it is known as dysgeusia, metallogueusia or cacogeusia (Zonneveld, 2011).

Around mid 2009, the NSW Food Authority began to receive calls to its Helpline from consumers experiencing pine mouth symptoms. Since these initial reports, the Authority has been keeping a watching brief on international developments. This issues paper details the information collected to date, summarising the most recent research on the cause of the pine mouth taste disturbance and recommends possible further activities for the food regulatory jurisdictions in Australia.

1.2 Symptoms

Accounts from sufferers of pine mouth indicate there is nothing at the time of consumption to suggest a difference with the pine nuts, they do not taste any different to 'normal' pine nuts. However, between 1 to 3 days after consuming the pine nuts the symptoms of the taste disturbance become evident and are usually described as a bitter or metallic taste which is exacerbated by the consumption of food and drink. The symptoms can last from a few days to up 2 weeks, although there are several anecdotal reports of symptoms lasting longer (up to 6-9 weeks) (Hampton, 2011; Tan, 2011). Prolonged duration of symptoms may possibly be due to ongoing consumption of pine nuts, with people not realising the cause of the taste disturbance.

While pine nuts are potentially allergenic, for all of the reported cases of pine mouth, the taste disturbance is not due to an allergy, and symptoms are self-limiting with no long-term adverse health effects observed (Ballin, 2012). As such, while pine mouth is not considered a food safety issue in the traditional sense, reports from consumers suggest that the taste disturbance is quite pronounced, significantly decreasing appetite and enjoyment of food and causing considerable discomfort to the sufferer. There are also anecdotal reports of sufferers undergoing unnecessary medical scans (eg MRI scans and endoscopy) or being prescribed medication (eg antibiotics) because there is insufficient knowledge of the condition among the population and the medical profession to realise the cause (Tan, 2011).

The degree of susceptibility to pine mouth appears to vary among people, as there are reports that consumers who eat pine nuts from the same batch may or may not experience the taste disturbance. Flesch (2011) and Ballin (2012) reported that females may be more frequently affected by pine mouth than males. Zonneveld (2011) found that it was necessary to consume at least six nuts (seeds) to bring on the bitter aftertaste, while Tan (2011) found that 2-3 seeds were enough to bring on symptoms. Ballin (2012) fed 6-8 pine nuts to volunteers and found that four out of six people developed classical symptoms. The severity of the symptoms may be dependent on the sensitivity of the person and the amount of pine

nuts consumed. Roasting pine nuts, or processing as in pesto, does not appear to make a difference and may still trigger the symptoms.

Tan (2011) found that approximately 35% of cases also experienced other non-taste symptoms such as headache, throat and stomach discomfort, nausea and bowel disturbance.

1.3 Increasing reports internationally

While first documented in 2001, the documented incidents of pine mouth since that time have been relatively rare. However, since 2009 there would appear to have been a significant increase in the number of reports occurring internationally. Contributing factors may include the increasing popularity of pine nuts in dishes such as pesto and gourmet salads and the sharing of information via the internet by consumers allowing people to more readily recognise the symptoms of pine mouth. However, the most significant factor may be the entry onto the world market of newer (non-traditional) species of pine nuts.

2. Pine nut production

2.1 Edible species of pine nuts

Pine nuts are the edible seeds of pine trees of the family *Pinaceae* genus *Pinus*. When first extracted from the pine cone, pine nuts are covered with a hard shell (seed coat), with the thickness dependent on the pine species. Pine nuts are not a true nut as (being a gymnosperm) they lack a carpel (fruit) outside. Unshelled pine nuts have a long shelf life if kept dry and refrigerated, but the shell must be removed before the nut is eaten. Shelled nuts can deteriorate rapidly in warm conditions, becoming rancid within a few weeks.

While there are greater than 100 species in the genus *Pinus*, there are around 29 different species listed by the Food and Agricultural Organization (FAO) as being cultivated for human consumption (Table 1 from FAO, 1998). Pine nuts are not commercially harvested in Australia or New Zealand and are predominantly imported from China, Spain, Italy and Turkey. Worldwide, the principal producing countries for pine nuts are Spain, Italy, Turkey, Portugal and China. Between January and November 2011, Chinese exports of pine nut kernels totalled 8,336 metric tons, of which about 45% was exported to the European Union (INC, 2011), with prices reaching up to \$US 25,000 per ton (INC, 2012).

In Asia, the two species most widely harvested are the Korean pine (*P. koraiensis*) which is the most important species in international trade and Chilgoza pine (*P. gerardiana*). The China Chamber of Commerce of Imports/Exports of Foodstuffs, Native Produce, and Animal By-Products (CCCFNA, 2011) indicate that *P. koraiensis* accounts for about 60% of the Chinese pine nut export share, followed by *P. sibirica* (Siberian stone pine – 12%), *P. yunnanensis* (Yunnan pine – 9%), *P. armandii* (Armand pine/Chinese white pine – 8%, used as a food ingredient only after roasting for paste), *P. pumila* (Japanese stone pine – 5%), *P. tabuliformis* (Chinese pine – 3%), *P. griffithii* (Himalayan pine – 1%) and *P. massoniana* (Masson pine – bird feed, inedible) – (Table 2). These varieties differ in origin, shape, size, colour, and taste – (Figure 1). Interestingly, not all of the above species classified as edible by the CCCFNA are included on the FAO list of edible species.

In Europe and the Middle East, the Stone pine (*P. pinea*) is most widely used and has a long history of cultivation dating back over 2,000 years (FAO, 1995). European pine nuts may be

distinguished from Asian ones by their greater length in comparison to girth. Asian pine nuts tend to be stubbier and shaped somewhat like long kernels of corn (Figure 1). Nuts of different pine species differ in size, nutritional value and taste however they may be mixed together at wholesale level and labelled generally as pine nuts (Fardin-Kia et al, 2012).

Table 1. Pine species with edible nuts

Species	Growing area	Remarks	
<i>P. pinea</i>	Spain, Portugal, France, Italy, Albania, Greece, Turkey	Important in international trade	
<i>P. gerardiana</i>	Afghanistan, Pakistan, India, Tibet		
<i>P. koraiensis</i>	Russia, China, Korea and Japan		
<i>P. sibirica</i>	Russia, Mongolia, Kazakhstan	Nuts are ground into cooking oil	
<i>P. edulis</i>	USA and Mexico	Important local producer of pine nuts	
<i>P. monophylla</i>	USA and Mexico		
<i>P. catarinae</i>	Mexico	Locally important	
<i>P. cembra</i>	Europe		
<i>P. cembroides</i>	Mexico		
<i>P. culminicola</i>	Mexico		
<i>P. juarezensis</i>	USA and Mexico		
<i>P. pumila</i>	Russia, China, Korea and Japan		
<i>P. quadrifolia</i>	USA and Mexico		
<i>P. albicaulis</i>	Canada and USA		Traditional food for indigenous tribes
<i>P. ayacahuite</i>	Mexico, Central America		
<i>P. coulteri</i>	USA		
<i>P. flexilis</i>	Canada and USA		
<i>P. lambertiana</i>	USA		
<i>P. ponderosa</i>	Canada and USA		
<i>P. sabiniana</i>	USA		
<i>P. strobiformis</i>	USA and Mexico		
<i>P. torreyana</i>	USA		
<i>P. johannis</i>	Mexico	Edible seeds	
<i>P. nelsonii</i>	Mexico		
<i>P. pinceana</i>	Mexico		
<i>P. remota</i>	USA and Mexico		
<i>P. lagunae</i>	Mexico	Edible seeds - found in single area	
<i>P. maximartinezii</i>	Mexico	Edible seeds - extremely rare	
<i>P. roxburghii</i>	India	Traditional food source	

Adapted from FAO (1998)

Table 2. Chinese pine nut varieties

<u>Chinese Name:</u>	<u>English Name:</u>	<u>Botanical Name:</u>
1) 红松	Korean Pine	<i>Pinus Koraiensis</i> * (ex Red Pine)
2) 西伯利亚红松	Siberian Stone Pine	<i>Pinus Sibirica</i> * (ex Cedar Pine or Snow Pine)
3) 云南松	Yunnan Pine	<i>Pinus Yunnanensis</i> *
4) 华山松	Huashan-Armand Pine	<i>Pinus Armandi</i> (to be avoided)
5) 偃松	Japanese Stone Pine	<i>Pinus Pimila</i> *
6) 油松	Chinese Pine	<i>Pinus Tabulaeformis</i> **
7) 马尾松	Masson Pine	<i>Pinus Massoniana</i> ** (ex Horse-Tail)
8) 乔松	Himalayan Pine	<i>Pinus Griffithii</i> *

* Suitable variety for direct human consumption.
 ** Edible, destined for food manufacturers.

Adapted from CCCFNA (2011)

Figure 1. Pine nut varieties (from Zonneveld, 2011)



Pine nut production can often fall short of demand, with the world total production estimated to be 20,000 tons of kernels in 2004 (Sharashkin & Gold, 2004). Harvests occur every two years and the quantity and quality of the harvest can fluctuate widely from year to year. The world market is often out of stock for months before the new harvest which has two major implications, high prices and price inelasticity (Sharashkin & Gold, 2004). Pine nuts are one of the most expensive nuts on the market, with retail prices of shelled nuts ranging up to \$66 per kg (retail price \$4.59 for a 70g package) and pine nut oil from \$60 (Guenard brand \$15.30 for 250mL) to \$230 per litre (Siberian Tiger Natural brand \$58 for 250mL)¹.

In 2009 there was a poor harvest of pine nuts in China, followed in 2010 by a poor North American harvest. As a result, during 2010-11 up to 80-90% of pine nuts sold in the USA were imported from China, which coincided with the first reports of pine nut syndrome in the USA (Zonneveld, 2011).

2.2 Non-edible species of pine nuts

The United Nations Economic Commission for Europe (UNECE, 1993) lists grades of pine nuts as 'Extra', 'Class I' and 'Class II', with nuts being downgraded due to defects in shape, development, colour, rancidity, mould or excess moisture.

Several species of pine nuts are considered not suitable for human consumption for a variety of factors, including *P. discolor* as they are too small and very hard and the species (Chinese red pine) (FAO, 1998; Destailats et al, 2010). However, the CCCFNA (2011) lists *P. armandii* as suitable for use as a food ingredient after roasting for paste. Many of the investigations into the cause of pine mouth have focussed on the *P. armandii* variety, especially those coming from growing areas of Shaanxi and Shanxi where the shell is thicker.

2.3 Pine nut oil and by-products

Pine nuts contain up to 68% oil and can be pressed to extract the oil, which is valued both for its mild, nutty flavour and its purported health benefits such as anti-inflammatory and antioxidant action (Sharashkin & Gold, 2004; Miraliakbari & Shahidi, 2008). Most commonly, *P. sibirica* and lower grade seeds of *P. yunnanensis*, and *P. koraiensis* are pressed commercially for the production of cooking oil which contains pinolenic, linoloneic and linoleic acids as well as polyunsaturated fatty acids. Apart from cooking and medicinal purposes, pine nut oil is used in cosmetics, beauty products and massage oil. The by-product of pine nut oil pressing is pine nut flakes, which are used in granolas, chocolates and crunch bars. These flakes contain up to 30% oil and when further pressed and crushed to extract this oil results in pine nut meal or flour which has a wide variety of culinary uses. It can be used as a gourmet substitute for wheat or rye flour, used in pastries and pancakes giving them a rich nutty flavour. When mixed with water, the pine nut meal becomes a pine nut milk or cream, which can impart a rich nutty flavour to dairy-free milk-like drinks (Sharashkin & Gold, 2004). To date there has been no reports linking pine nut oil or other by-products to the pine mouth taste disturbance.

¹ Prices correct as at December 2011

3. Reports of pine mouth

3.1 Reports of pine mouth in Australia

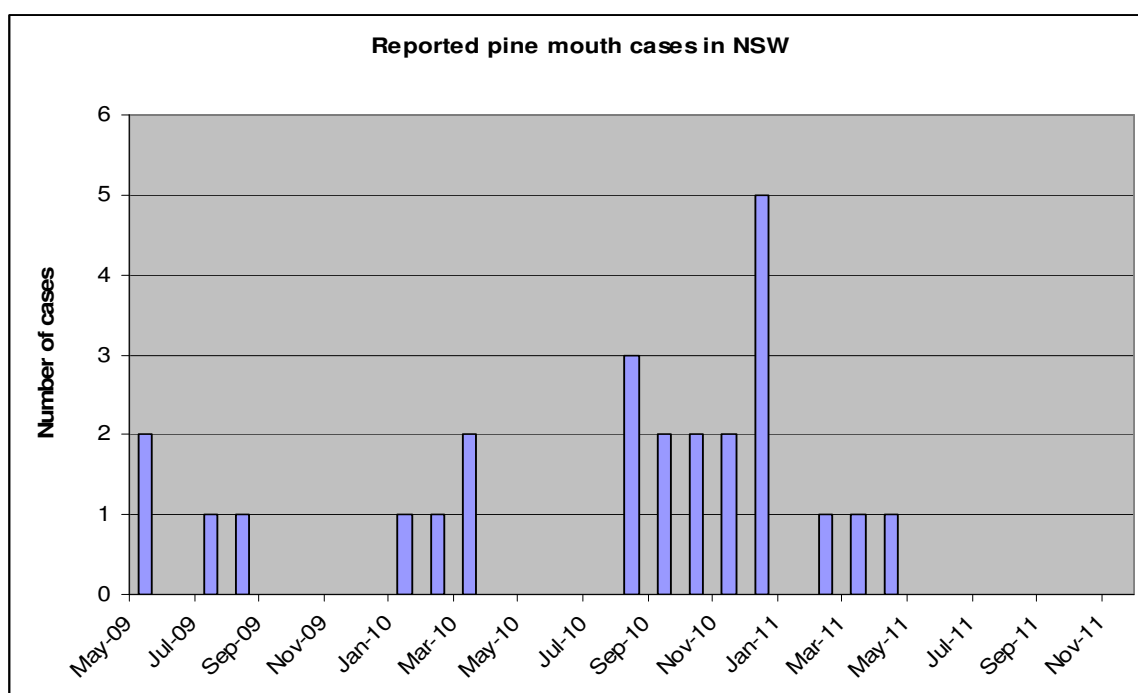
Initial informal discussion with Food Standards Australia New Zealand (FSANZ) has indicated they have received a few calls concerning pine mouth. FSANZ has a fact sheet on pine nuts and the pine mouth taste disturbance on its website (FSANZ, 2010) and has responded to around 10-15 enquiries regarding 'pine mouth' since 2009. A survey of Australian food regulatory jurisdictions has found that in:

- DAFF Biosecurity - one case reported through to during early 2012
- Queensland - one case was reported during 2010, affecting two people
- Tasmania - two cases reported in 2010, one additional case reported in 2011 who contacted NSW
- Victoria - three cases reported
- NSW - the first report of pine mouth came through to the NSW Food Authority in May 2009. Since that time, the Authority has received 26 calls regarding bitter/metallic taste associated with pine nuts, eighteen of those calls were received in 2010 (Figure 2). One call was received in July 2012.

No single brand has been implicated in the calls received, with complainants mentioning several brand names and retailers.

Many of the people contacting the Authority had already searched the internet and found information on pine mouth and attribute their symptoms to the consumption of imported pine nuts, particularly those imported from China. The complainants have generally sought for the Authority to further investigate the problem.

Figure 2. Reported cases of pine mouth in NSW (to November 2011)



3.2 International reports

Since the initial literature report in 2001, there have been several reports from Europe in the UK, Ireland, France, Germany, Switzerland and the USA documenting the pine mouth taste disturbance (UKFSA, 2009; FSAI, 2009; Flesch, 2011; BFR, 2011; FDA, 2011). A blog created by a Masters research student in the Netherlands claimed that they had received 434 reports of pine nut syndrome from 23 countries (Tan, 2011).

France

In September 2009, the French Food Safety Agency (AFSSA, 2009) issued a paper concerning the bitter taste associated with pine nut consumption. In the paper the agency noted there has been over 800 reports of the taste disturbance in France and appeared to be related to the consumption of Korean pine (*P. koraiensis*) grown in China and Pakistan. At the time, investigations were ongoing and testing did not reveal any contaminants, either chemical or biological. Since most other reports of pine mouth do not implicate *P. koraiensis* and it is a variety that is widely consumed, it is concluded that the French may have incorrectly implicated *P. koraiensis*, and instead it may have been a mixed batch including *P. armandii*.

The subsequent follow-up report in July 2010 by the French Agency for Food, Environmental and Occupational Health Safety (ANSES, 2010) did not implicate *P. koraiensis* as a potential cause, but noted that not all pine nuts being sold were listed as edible by the FAO, including *P. armandii*, *P. massoniana*, *P. yunnanensis* and *P. tabuliformis* (FAO, 1998). The agency noted that these species appear to be grown in China and marketed locally as food, however there may have been mixing of edible pine nuts with species not typically used for human consumption. The agency suggested undertaking different analyses to determine the species of pine nuts being sold (noting that some products contain mixed species of pine nuts) and restricting consumption of pine nuts to only those listed by the FAO (1998).

More recently, Flesch et al (2011) published a paper documenting more than 3000 cases of pine mouth in France reported between March 2008² and January 2010. The number of cases rose sharply from May 2009 to reach a peak in August 2009 with 697 cases. The median time to onset of symptoms was 24 hours and it lasted less than 14 days in 95% of cases. Raw as well as cooked or processed pine nuts were implicated.

United Kingdom

In July 2009, the UK Food Standards Agency (UKFSA) issued a news release regarding pine mouth (UKFSA, 2009). In this release they stated that they had received various reports regarding pine mouth and encouraged people to contact the agency to report cases.

A subsequent news article in 2011 quoted that the Food Standards Agency was investigating hundreds of cases of pine mouth, with between 10 and 15 cases of pine mouth reported every month in Britain since 2009 (Jamieson, 2011). The Agency itself indicated that it had received a steady flow of reports since 2009 and now has over 500 complaints. The reports showed that 99% of cases were linked to pine nuts from China (UKFSA, 2011a).

² The time of the first calls through to the French poison centres reporting persistent bitterness following the ingestion of pine nuts

Hampton et al (2011) documented a case in a letter to the *British Dental Journal* involving a 47-year-old male who presented reporting a metallic taste (metalloguesia) and a soapy/woolly sensation to the tongue and lips which persisted for six weeks.

Ireland

In September 2009, the Food Safety Authority of Ireland (FSAI) also issued an alert regarding pine mouth (FSAI, 2009). In their alert they associated the taste disturbance to pine nuts imported from China and were to continue monitoring reports of cases.

United States

A case of pine mouth was described in the *Journal of Medical Toxicology* (Munk, 2010) with the suggestion that pine nuts imported from China into the US were responsible for the taste disturbance. The US Food and Drug Administration (FDA) placed a media release on their website in 2011 regarding pine mouth and the consumption of pine nuts (FDA, 2011). The FDA developed a detailed questionnaire, and collected and analysed samples from some consumers submitting complaints. The Agency found that the majority of pine nuts associated with the pine mouth taste disturbance were eaten in the raw state (either as snacks or as a component of salad or pesto sauce). It also found that consumers did not detect a rancid or off-taste when eating the pine nuts.

The FDA has continued to analyse consumer complaints to determine whether the severity of symptoms and likelihood of developing them is related to the amount of pine nuts consumed. Between July 2008 to June 2011, FDA field offices received 411 complaints from consumers reporting symptoms consistent with pine mouth or pine nut syndrome (Handy et al, 2011).

Switzerland

A case of pine mouth in Switzerland was described in the *American Journal of Medicine* by Picard & Landis (2010). Symptoms detailed in this case study were similar to the typical symptoms described previously, with the author suggesting that pine nuts being imported from Asia as being the cause of the taste disturbance.

The Netherlands

The blog associated with the Masters thesis of Tan (2011) surveyed complaints across a large number of countries, including 58 cases of pine mouth in the Netherlands.

3.3 Reports of pine mouth in the media and social media

Reports of the pine mouth taste disturbance have appeared in the press in the US and UK (Hutchison, 2010; Middleton, 2009; Jamieson, 2011; Daily Mail Reporter, 2011). Jamieson (2011) reported in the UK's *The Telegraph* in April 2011 that "cheap Chinese exports of pine nuts have been blamed for a rare mouth condition that leaves a bitter aftertaste for weeks". The newspaper reported that UK supermarkets "have now pledged to import only from approved suppliers after customers complained of a foul, metallic taste in their mouths". Reports indicate that some of the major supermarket chains in the UK such as Sainsbury's may have changed their purchasing policy with regards to pine nuts and only used approved exporters, or have put in place measures to ensure that batches of pine nuts do not include *P. armandii*.

Pine mouth has also been widely discussed on internet forums (Park, 2009; The Body Odd, 2010), online blogs by a British botanist (Hyam, 2008), a Masters student (Tan, 2011), and an Australian blog (FNQhome.com, 2009) as well as the English consumer interest group 'Which?' (2011). In addition, a Facebook support group for sufferers of pine mouth has been established (Facebook, 2011). While the numbers of reported cases in most countries still appear relatively small, the number of unreported cases may be much larger given the number of anecdotal responses and comments that typically appear in response to these reports and blogs. For instance, in response to a post in Britain by Hyam (2008) there were 1,515 responses up to May 2010, while in Australia the FNQhome.com blog (2009) received 72 comments between July 2009 and December 2011.

4. Research into causes of pine mouth

4.1 Pine nuts responsible for causing pine mouth

In the initial report of pine mouth from Mostin (2001), the author did not record the species of the pine nuts, but stated they were imported from China. Since that time, based on the published reports of pine mouth, Destailats et al (2010) concluded that there appears to be a growing correlation between the taste disturbance and consumption of *P. armandii* (Chinese white pine) and *P. massoniana* (Chinese red pine).

While there are suspicions about a particular species of pine nut being responsible for the taste disturbance, there have been several obstacles to overcome in making a definitive conclusion. The lack of methodology to accurately distinguish pine nut species (especially when they are sold in mixed batches) has been a primary difficulty and researchers have in some cases resorted to sorting pine nuts based on visual classification or attempting to classify on the basis of fatty acid profile. In addition, there is still incomplete understanding of the mechanism for what causes the pine mouth symptoms in susceptible consumers. Nevertheless, in a recent journal article Zonneveld (2011) reported that tasting showed definitively that *P. armandii* is the origin of the bitter aftertaste known as pine nut syndrome.

4.2 Methods of analysis for pine nut speciation

In an effort to assist with the research and investigation into pine mouth, Destailats et al, (2010) published a method for the speciation of pine nuts based on their fatty acid profile using gas-liquid chromatography. This method involves extracting the lipids from the pine nuts followed by gas-liquid chromatography (GLC) analysis. The percentage contribution of each fatty acid is then calculated and used to determine a diagnostic index (DI). The DI value for a sample of pine nuts is then compared against known values for the different *Pinus* species and has been shown to successfully differentiate between different pine nut species. During their analysis, Destailats et al, (2010) found *P. armandii* and *P. massoniana* in commercial samples, and noted they were not included in the list of edible species by the FAO (1998). The distinctive feature of the fatty acid profile for *P. armandii* seeds is the lower level of octadecenoic acids and the higher level of taxoleic acid compared to other *Pinus* species (Destailats et al, 2010). However, it has been noted that for some commercial samples where there are mixed pine nuts species present, separation of pine nuts based on morphology is needed before analysis (Destailats et al, 2010; Fardin-Kia et al, 2012).

In France, the method of Destailats et al, (2010) was proposed to assist in further investigating cases of pine mouth (ANSES, 2010). Follow-up work by Destailats et al, (2011) on 16 pine nut samples from consumers reporting pine mouth symptoms in Belgium (n = 5), France (n = 1), Netherlands (n = 8), New Zealand (n = 1) and one sample where the origin was not listed. Using the same GLC methodology previously described, the authors confirmed the presence of *P. armandii* in all samples, either pure (n = 12) or in mixture with *P. koraiensis* (n = 4).

Köbler et al (2011) used nuclear magnetic resonance (NMR) spectroscopy and chemometrics on 57 samples of pine nuts as a screening method to identify the species causing pine mouth. Samples could be classified into either three groups (based on ¹H NMR) or two groups (based on ¹³C NMR). The authors found that *P. armandii*, *P. massoniana* and *P. koraiensis* were categorised into one group, while *P. pinea* and *P. gerardiana* were categorised into either one or two separate groups depending on the methodology. Since the first group contains both species suspected of causing pine mouth and *P. koraiensis* which is traded internationally, further discrimination would be required.

Similarly, Handy et al (2011) examined 15 samples of pine nuts implicated in cases of pine mouth by DNA-based identification of the chloroplast *ycf1* gene. They found the methods of both Destailats et al (2010) and Köbler et al (2011) were not specific enough for identification of separate *Pinus* species when present in a mixed batch of pine nuts. Of the 15 samples Handy et al (2011) examined, 11 contained mixed species of pine nuts and all contained *P. armandii*, despite the fact that the International Nut and Dried Fruit Council (INC) maintains this species is only exported to the USA for use in manufacturing paste. The authors proposed their method could be used by regulators to distinguish whether *P. armandii* was absent or present in a sample, even when mixed with other pine nuts.

Zonneveld (2011) used genome size, as measured by nuclear DNA flow cytometry, to identify nut species. The author reported that subsequent taste testing showed 'definitively' that *P. armandii* is the origin of the bitter aftertaste known as pine nut syndrome.

The International Nut and Dried Fruit Council (INC) have also commissioned an analysis of different species of pine nuts and profiled their fatty acid composition. Although initially due in December 2010 (INC, 2010), a report regarding these results has not yet been published.

4.3 Proposed mechanism of pine mouth

The causative agent responsible for the pine mouth taste disturbance has so far not been identified. Several mechanisms have been proposed since the original report of pine mouth where Mostin (2001) noted that the implicated pine nuts were oxidised. No fungal contamination was observed and an analysis of the samples did not detect any pesticides or heavy metals. The implicated pine nuts were examined chromatographically against other pine nuts. This resulted in the isolation of triglycerides formed by C16-18 unsaturated fatty acids from the bitter tasting batches. Based on this, it was hypothesised by the author that these triglycerides may play a role in the taste disturbances. However, subsequent publications have discounted oxidative reactions as the cause of pine mouth, as the delayed onset and duration of symptoms is usually too long to be explained by rancidity (Möller, 2010; Köbler et al, 2011; Zonneveld, 2011). In addition, studies of the oxidation of pine nut oil by Miraliakbari & Shahidi (2008) show that its properties are not significantly different enough from other nut oils to explain pine mouth symptoms (Möller, 2010).

Since 2001, most activity and research in relation to pine mouth appears to be occurring in Europe, with France and the Netherlands most proactive in attempting to determine the cause of pine mouth. Destailats al (2010) presented a hypothesis that the pine mouth symptoms may be due to an unidentified toxin present in some varieties of non-edible pine nuts. It was thought that the delayed onset and persistence of the taste disturbance were due to a toxin that acts via an unknown toxic mechanism on the receptor. The authors stated that more experimental work was necessary through continued monitoring of poison cases, botanical and biochemical analysis, and experimental studies.

Blog 'The Body Odd' (2010) quoted Dr Alan Hirsch, from the Smell & Taste Treatment and Research Foundation in Chicago as saying "At this moment, we think it's the sweet taste receptors that aren't firing off. In response to the lack of the taste of sweet, bitter comes out more."

Zonneveld (2011) suggests the explanation proposed by Möller (2010) may be most plausible. Pinolenic acid in pine nuts stimulates the endocrine system to produce the hormone cholecystokinin (CCK) in the upper intestine (Hughes et al, 2008). Pasman et al (2008) found that the pine nut oil from the Korean pine nut (*P. koraiensis*) was eight-fold more potent in releasing CCK than Italian stone pine nut oil (*P. pinea*). The release of CCK is valuable for appetite suppression and stimulates the production of digestive enzymes. This slows the emptying of the stomach and causes a contraction of the gall bladder to release bile in the duodenum and also induces the liver to produce more bile. Furthermore, there are bitter taste receptors present in both the gastrointestinal tract as well as in the mouth. Möller (2010) proposed that the production of excess bile can cause a cross wiring of the brain's neurological bitter sensation of the tongue and gastrointestinal tract, leading to the metallic or bitter aftertaste similar to that experienced by sufferers of pine mouth.

However, the duration of pine mouth symptoms exceed the normal digestive cycle residence times. To explain this, Möller (2010) examined the nature of the digestive system and explains how one of the processes the liver uses to remove waste products and metabolites is through the bile ducts, allowing the gall bladder to release processed metabolites back into the digestive system in the duodenum (the first part of the small intestine) for faecal elimination. As such, some metabolites processed by the liver can then be reabsorbed lower in the small intestine and returned to the liver via the portal vein, through a process called enterohepatic recirculation (EHR). This form of bile reflux can extend the residence time in the gut and can explain why it sometimes takes more than one week for the bitter taste disturbance to disappear and why it takes one or two days for the symptoms to appear. It might also explain the occasionally reported cases of diarrhoea, nausea, and abdominal cramping (Zonneveld, 2011).

At this point in time, no mechanism for pine mouth has been verified and no single compound has been isolated from pine nuts that can be conclusively pinpointed as the cause for the pine mouth taste disturbance. As summarised by Köbler et al (2011), the substance could be a naturally occurring component of the seeds or could represent a contaminant or intentional addition during harvesting or processing of the seeds (shell removal, drying, preservation, exposure to light and air etc.).

5. Action to date

5.1 European Commission

At a meeting in January 2011, the Standing Committee on the Food Chain and Animal Health (SCFAH) for the European Commission confirmed that, batches of pine nuts containing the species *P. armandii* were considered to be unfit for human consumption (European Commission, 2011). They were therefore deemed to be unsafe in accordance with Article 14 of Regulation (EC) 178/2002 laying down the general principles and requirements of food law.

Several European countries have recalled pine nuts due to the taste disturbance phenomena. Reports submitted to the European Commission's Rapid Alert System for Food and Feed (RASFF) shows 37 notifications of 'taste disturbance' associated with pine nuts during 2011, with China being the main origin. Many of these pine nut batches appear to have been imported from China through the Netherlands and Germany (Appendix 3). The Czech Agriculture and Food Inspection Authority (CAFIA) in particular have been very active in withdrawing product from the market due to consumer complaints of taste disturbance, although it states that some product originated in Pakistan, as well as China (CAFIA, 2011). It is unclear whether this was due to blending of products from these two countries.

Further, in France the ANSES (2010) recommended action in relation to pine mouth includes:

- verifying that only the species listed as edible by the FAO of the United Nations are imported
- continue to monitor the number and nature of complaints by consumers
- visually inspecting pine nuts, in particular seeking out nuts that are of a small size
- evaluate the usefulness of published methods for verifying pine nut species

On the 15th September 2010, the French Ministry of Economy published in the Official Journal of the French Republic an opinion intended for pine nuts producers, importers and distributors, stating that the pine nut species *Pinus armandii* and *Pinus massoniana* fall in the scope of Regulation 258/97 concerning novel foods and novel food ingredients (cited in Anon, 2010). Novel foods are defined as having no history of 'significant' consumption in the European Union prior to 15 May 1997 and must be authorised according to the Novel Food legislation. In the absence of an authorisation, such products cannot be marketed. Although pine nuts generally have a long history of safe consumption and therefore could not be considered a novel food, the French government have argued that these particular species of pine nut do not have a safe history of consumption and have not traditionally been consumed. To date, no other European country seems to have taken this approach.

5.2 Codex Alimentarius Commission

At the session of the Codex Committee on Pesticide Residues (CCPR) held in Beijing, China during April 2011, the classification of Tree Nuts was amended to exclude *P. armandii* and *P. massoniana* from the list of edible tree nuts. The basis for this decision was that the Committee agreed these pine nut species have a bitter taste and are not fit for consumption (Codex Alimentarius Commission, 2011).

5.3 Measures implemented by businesses importing and selling pine nuts

All pine nuts are imported into Australia as there is no local commercial harvest. However they are normally labelled as 'Packaged in Australia from imported ingredients' with no specific country of origin noted and there is no labelling with the botanical name. The main brands of pine nuts sold in Australia can be generally found in health food stores, supermarkets and green grocers (Appendix 1).

In November 2010, a major Australian retailer sent a letter to their suppliers giving instructions that the *P. armandii* and *P. tabuliformis* were to be excluded from their pine nut supply. The retailer also required documented traceability for the species of pine nuts sources (including scientific name) to the origin of harvest. Information from the Food & Beverage Importers Association of Australia (Beaver *pers comm*) indicates that the three main importers of pine nuts into Australia use accredited pine nut exporters and specify the variety *P. koraiensis* as the required type. Visual checks are conducted on all deliveries with the kernel size verified, as *P. koraiensis* as this variety has a smaller kernel size 550-750 kernels/100g compared with 850-1500 kernels/100g for *P. armandii* (CCCFNA, 2011).

In a response to a blog from FNQhome.com (2009), the General Manager Sales and Marketing Richard Genest of Stahmann Farms, which recently purchased the Ducks nuts brand, wrote to assure customers that the company has acted to ensure that they only import and package *P. koraiensis* pine nuts.

Some of the major supermarkets in the UK are now only using suppliers of pine nuts from an approved list compiled by the CCCFNA (INC, 2011c). However, in April 2011, a UK consumer group, 'Which?' reported that, despite being told that measures had been introduced to prevent pine mouth from happening, judging by the volume of correspondence they were still receiving from people, "it's clear the problem persists" (Which?, 2011).

5.4 Industry measures implemented by Chinese exporters

The CCCFNA and the Chinese Tree Nuts Association met in December 2010 to discuss the pine nut taste disturbance issue. While the species *P. armandii* is the prime suspect as causing pine mouth, there are up to five official varieties of *P. armandii* and it may only be one thick-shelled variety originating from the regions of Shaanxi and Shanxi in China that is responsible for causing the taste disturbance and was not suitable for human consumption (INC, 2011; Tan, 2011). Other growing areas where *P. armandii* grows with a thinner shell were not considered problematic and have a normal taste. However, the CCCFNA in co-operation with the Chinese authorities had decided to implement measures to minimise the occurrence of pine mouth by not exporting any *P. armandii*.

The INC announced these measures through a media release that measures had been strengthened in China to avoid the mixing of edible and inedible pine nut species *P. armandii* and a list of reliable accredited Chinese exporters of pine nuts had been established which came into effect in July 2011 (Appendix 2). The Chinese industry has reminded pine nut exporters not to export *P. armandii* to the EU market for food purposes and the CCCFNA has undertaken to make checks on pine nut shipments. No mention was made in the media release of other export markets (INC, 2011a, INC, 2011b, INC, 2011c).

The new measures are summarised on the UKFSA website (UKFSA, 2011b) as including:

- All pine nut exporters should be accredited members of CCCFNA
- The processing plants must be accredited for HACCP and ISO9002 or ISO20002 and have certification of "Enterprises for Food Export Registration" from the Chinese Inspection and Quarantine Service (CIQ).
- All exported pine kernels must be compulsorily inspected by SGS as a third party with particular attention to the "pine mouth" problem
- The outer packaging of exported pine nuts should clearly show the name of the pine kernels in Chinese, English and the botanical name, sizes, batch number, name of processor for full traceability.
- Consignments of pine nuts from China should be accompanied by a Certificate of Export and an Inspection Certificate and Pine nut exporters should be members of CCCFNA. The certificates should be signed by an authorised SGS inspector.
- The United Nations Economic Commission for Europe (UNECE) specifications for pine nut kernel must be adhered to
- Any notification of 'taste disturbance' of pine kernels imported from China will be communicated from DG SANCO directly to CCCFNA through the official channels.
- For any non-complying Chinese processor or exporter that CCCFNA will take the necessary steps to radiate the company from the approved list of suppliers of the pine nut export group.

In addition, the CCCFNA have released the third issue of the Chinese Pine Nut Handbook (CCCFNA, 2011). This handbook acknowledges that some pine nuts are good for food, some are best used as an ingredient and some should only be used for oil extraction.

Despite the introduction of these additional measures in mid-2011, the number of RASFF notifications for taste disturbance due to pine nuts in Europe dropped only slightly from 21 during the first half of 2011 to 16 during the second half. It was acknowledged that the effects of the new rules may not be fully seen until early 2012, because the shelf life of pine nuts (up to two years) may mean that batches exported in late 2010 or early 2011 may still cause problems (INC, 2012). To date there have only been two RASFF notifications in January and February 2012 which may indicate the measures are having the desired effect.

The emphasis of measures introduced in China was to ensure that unsuitable pine nuts were not sent to the European Union, and at this stage similar guarantees have not been obtained for exports of pine nuts from China to Australia. The Australian Food & Beverage Importers Association is attempting to obtain similar undertakings for pine nut exports to Australia (Beaver *pers comm*).

6. Conclusion

Worldwide there has been a significant increase in the notifications of pine mouth since 2009. Although the cause of the taste disturbance has not been conclusively resolved, the evidence continues to mount that mixing species of pine nuts traditionally regarded as inedible (*P. armandii* and *P. massoniana*) with edible pine nuts may be a significant contributing factor.

As noted by the French government, because *P. armandii* is not listed as edible by the FAO, and appears to be implicated as one of the varieties that more commonly causes pine mouth, then excluding this species would be the first step in reducing the risk of exposure to consumers. This is evident with Codex categorising certain species of pine nuts as unfit for human consumption and European countries rejecting shipments of pine nuts due to the taste disturbance.

The Chinese authorities have implemented new measures on pine nut exporters to minimise the presence of *P. armandii* in shipments to the European Union. Since the introduction of these measures, the number of border rejections for pine nuts in the EU appear to be declining, which may indicate the measures are having an effect.

Recommendations

The following are recommendations made for Australian food regulatory authorities to minimise the future occurrence of pine mouth cases:

- i. *Evaluate the ongoing effectiveness of industry measures introduced in China.*

The total number of reported cases in Australia is relatively small (~30 cases) although it is acknowledged that the actual number may be significantly more, based on blog comments. Despite the unpleasant symptoms of pine mouth, the lack of long-term harm to consumers means the pine nuts may not necessarily be considered 'injurious to health'.

Market forces have driven importers and retailers to ensure selection of appropriate pine nuts with the major Australian retailers specifying only *P. koraiensis* to be sold. The measures introduced in China should be taking effect now, however at this stage the same commitment that was made to the EU has not necessarily been made for pine nut exports from China to Australia.

- ii. *Continue to monitor any reports of pine mouth cases in Australian consumers and determine the source of any implicated pine nuts*

Should reports of pine mouth be made in the future, attempts should be made by regulatory authorities to discern the source and species of pine nut causing the taste disturbance to better inform future responses.

- iii. *If numbers of pine mouth cases significantly increase in Australia, refer to the Implementation Sub-Committee (ISC) for consideration and decision.*

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Appendix 1. List of approved Chinese pine nut kernels shippers

List taken from INC press release on 25 July, 2011 (INC, 2011c)

A) Heilongjiang Province:

- 1) Heilongjiang Hongtai Pine nut Co., Ltd.

B) Jilin Province and Meihekou City of Jilin Province:

- 2) Jilin City Changrong Agricultural & By-Products Corporation
- 3) Jilin Ruihua Foods, Co., Ltd.
- 4) Yanji Shoei Foods Co., Ltd.
- 5) Jilin Meihekou Tianyuan Green food Corporation
- 6) Jilin shengyuan Food Co. Ltd.
- 7) Jilin Zhongxing Foodstuff Holding Co., Ltd.
- 8) Jinlin Tianzhu Food Co., Ltd.
- 9) Li Feng Foodstuffs Co., Ltd of Meihekou City
- 10) Meihekou City Hewang Food Co., Ltd.
- 11) Meihekou city Sanhewanxin Mountain Specialty Co., Ltd.
- 12) Meihekou City songhai Special Products Co., Ltd.
- 13) Meihekou City Wanxing Food Co., Ltd.
- 14) Meihekou city Yunfeng Kernels Processing Co., Ltd.
- 15) Meihekou Fly Dragon Food Products Co., Ltd.
- 16) Meihekou Sea Food Co., Ltd.

C) Liaoning Province:

- 17) Dalian Jinyu Foods Co., Ltd.
- 18) Fuxin Zhenlong native Produce Ltd.
- 19) Golden pacific (Dalian) Food Co. Ltd.

D) Yunnan Province:

- 20) Kunming Xinwei Food Co., Ltd.

E) Shanxi Province:

- 21) Fen zhou Yu Yuan Native Produce Co., Ltd.
- 22) Taiyuan Tianhe Food Group Corporation Ltd.

F) Tianjian:

- 23) Tianjin Dinghao Nuts & Spices Import & Export Co., Ltd.
- 24) Tianjin native Produce Imp & Exp Group Corporation Ltd.
- 25) Sunshine (Tianjin) Produce Ltd.

G) Beijing:

- 26) Voicevale (Beijing) Trading Co., Ltd.

NOTE – a subsequent list published on the UKFSA website as at 11 April 2012 does not contain the businesses listed in C, D, E, F and G.
(http://www.food.gov.uk/foodindustry/imports/banned_restricted/pine-nuts-china/pine-nut-exporters)

Appendix 2. Notifications to European RASFF on pine nut taste disturbance

Date	Notification basis	Notified by	Subject	Action taken
10/03/2010	border control - consignment detained	ITALY	altered organoleptic characteristics of pine nuts from China	Border rejection, re-dispatch
23/08/2010	border control - consignment detained	GREECE	altered organoleptic characteristics of pine nuts from Turkey infested with moulds and with insects	Border rejection, re-dispatch
31/01/2011	consumer complaint	DENMARK	taste disturbance (bitter aftertaste) caused by organic pine nut kernels from China, via the Netherlands and via Germany	withdrawal from the market
3/02/2011	consumer complaint	BELGIUM	taste disturbance (bitter aftertaste) caused by pine nuts (<i>Pinus yunnanensis</i>) from China, via Germany and via the Netherlands	withdrawal from the market
18/02/2011	consumer complaint	DENMARK	taste disturbance (bitter aftertaste) caused by organic pine nut kernels from China, via the Netherlands	withdrawal from the market
18/02/2011	consumer complaint	CZECH REPUBLIC	unsuitable organoleptic characteristics (pine seeds: bitter taste; Brazil nuts: rancid taste) of and taste disturbance caused by nut mix from Slovakia infested with moulds	withdrawal from the market
7/03/2011	consumer complaint	CZECH REPUBLIC	taste disturbance caused by pine nuts from China, via the Netherlands	withdrawal from the market
8/03/2011	company's own check	DENMARK	pine nuts from the Netherlands, with raw material from China, packaged in Denmark unfit for human consumption (presence of <i>Pinus armandii</i>)	withdrawal from the market
18/03/2011	consumer complaint	MALTA	taste disturbance caused by pine nuts from China, packaged in Malta, via Germany and via Italy	withdrawal from the market
24/03/2011	consumer complaint	FINLAND	taste disturbance caused by pine nuts in salad mix with raw material from China, via Sweden	
29/03/2011	consumer complaint	IRELAND	taste disturbance caused by pine nuts from China	withdrawal from the market
19/04/2011	consumer complaint	DENMARK	taste disturbance caused by organic pine nut kernels from the Netherlands	withdrawal from the market
16/05/2011	consumer complaint	NORWAY	taste disturbance (mixture <i>Pinus sibirica</i> and <i>Pinus armandii</i>) caused by pine nuts from Norway, with raw material from China, via the Netherlands	withdrawal from the market
26/05/2011	consumer complaint	BELGIUM	taste disturbance (<i>Pinus armandii</i>) caused by pine nuts from Belgium, with raw material from China, via the Netherlands	recall from consumers
1/06/2011	consumer complaint	BELGIUM	taste disturbance caused by pine nuts from China, packaged in Romania, via the Netherlands	recall from consumers
15/06/2011	consumer complaint	MALTA	taste disturbance caused by pine nuts from China, packaged in Malta, via Italy	recall from consumers
16/06/2011	consumer complaint	MALTA	taste disturbance (<i>Pinus armandii</i>) caused by pine nuts from China	withdrawal from the market
17/06/2011	consumer complaint	IRELAND	taste disturbance caused by pine nuts from China, via the Netherlands	recall from consumers
17/06/2011	consumer complaint	DENMARK	taste disturbance (presence of <i>Pinus armandii</i>) caused by organic pine nuts from China, via Germany	
24/06/2011	consumer complaint	CZECH REPUBLIC	taste disturbance caused by pine nuts from China, via the Netherlands	withdrawal from the market

Date	Notification basis	Notified by	Subject	Action taken
24/06/2011	consumer complaint	CZECH REPUBLIC	taste disturbance caused by pine nuts from China, via the Netherlands	withdrawal from the market
29/06/2011	official control on the market	CZECH REPUBLIC	taste disturbance caused by pine nuts from China, packaged in the Czech Republic, via the Netherlands	withdrawal from the market
30/06/2011	consumer complaint	BELGIUM	taste disturbance (presence of <i>Pinus armandii</i>) caused by pine nuts from the Netherlands	recall from consumers
1/07/2011	official control on the market	DENMARK	taste disturbance caused by organic pine nuts from China, via the Netherlands	withdrawal from the market
18/07/2011	consumer complaint	BELGIUM	taste disturbance (presence of <i>Pinus armandii</i>) caused by pine nuts from China, via the Netherlands	official detention
3/08/2011	consumer complaint	BELGIUM	taste disturbance (presence of <i>Pinus armandii</i>) caused by pine nuts from the Netherlands	withdrawal from the market
10/08/2011	consumer complaint	BELGIUM	taste disturbance caused by pine nuts from China, via the Netherlands	withdrawal from the market
16/08/2011	consumer complaint	CZECH REPUBLIC	taste disturbance caused by pine nuts from China, packaged in Slovakia, via Germany	re-dispatch
9/09/2011	official control on the market	GERMANY	taste disturbance caused by pine nuts from China	no action taken
22/09/2011	consumer complaint	IRELAND	taste disturbance caused by pine nuts from the United Kingdom	withdrawal from the market
28/09/2011	consumer complaint	CZECH REPUBLIC	taste disturbance caused by pine nuts from China, via the Netherlands	withdrawal from the market
30/09/2011	consumer complaint	CZECH REPUBLIC	taste disturbance caused by pine nuts from Pakistan and China, via Germany and via Slovakia	withdrawal from the market
13/10/2011	consumer complaint	BELGIUM	taste disturbance (presence of <i>Pinus armandii</i>) caused by pine nuts from China, via the Netherlands	withdrawal from the market
24/10/2011	consumer complaint	CZECH REPUBLIC	taste disturbance caused by pine nuts from China, via the Netherlands	official detention
8/11/2011	consumer complaint	UNITED KINGDOM	taste disturbance caused by pine nuts from China, via the Netherlands	withdrawal from the market
9/11/2011	consumer complaint	IRELAND	taste disturbance caused by pine nuts from China, packaged in the United Kingdom, via the Netherlands	no action taken
9/11/2011	consumer complaint	IRELAND	taste disturbance caused by pine nuts from China, via Germany and via the United Kingdom	no action taken
16/11/2011	consumer complaint	IRELAND	taste disturbance caused by pine nuts from China, via the Netherlands and via the United Kingdom	no action taken
22/11/2011	consumer complaint	IRELAND	taste disturbance caused by pine nuts from China, via the Netherlands	no action taken
5/01/2012	consumer complaint	CZECH REPUBLIC	taste disturbance caused by pine nuts from China, via the Netherlands	withdrawal from the market
17/02/2012	consumer complaint	DENMARK	taste disturbance (<i>Pinus armandii</i>) caused by organic pine nuts from China, via Germany	Recall from consumers

adapted from RASFF (2012) as at 18 June 2012