Sveriges lantbruksuniversitet
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Department of Molecular Sciences

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Degree project • 15 credits
Agricultural Programme - Food Science
Molecular Sciences, 2019:26
Uppsala, 2019

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Credits:	15 credits
Level:	First cycle, G2E
Course title:	Independent project in Food Science
Course code:	EX0876
Programme/education:	Agricultural Programme - Food Science
Course coordinating department:	Department of Molecular Sciences
Place of publication:	Uppsala
Year of publication:	2019
Title of series:	Molecular Sciences
Part number:	2019:26
Online publication:	https://stud.epsilon.slu.se
Keywords:	oats, acrylamide, free asparagine

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Abstract

Acrylamide can be formed during heating of cereals. Oats have the second highest acrylamide forming potential of rye, wheat, barley and oats. Several studies have found high levels of acrylamide in oat products that exceed the benchmark limits within the EU. These limits might be strict limits in future which every food producer needs to follow.

Reactions between reducing sugars and free asparagine is the major contribution of acrylamide in food. Free asparagine is not incorporated into proteins and might thus easily react in the *Maillard* reaction and form acrylamide. It has been shown that free asparagine is the determining factor for acrylamide formation in cereals. Free asparagine accumulates in cereals when there is an excess of nitrogen and at the same time a deficit of sulphur. Proper fertilising and selection of crops with low levels of free asparagine might be the key to lower acrylamide formation in oat products.

During processing, salts, other amino acids than asparagine and asparaginase may be added to lower acrylamide formation in various baked products. Some of these additives and especially salts have however bad impacts on health as well as product quality. Acrylamide formation is also affected by pH, where a higher pH produces more acrylamide. Antioxidants might inhibit acrylamide formation, but some studies have found the opposite and theorises that some antioxidants prevent further reaction of acrylamide with other compounds. It is known however that antioxidants can inhibit lipid oxidation and by extension lower the formation of acrylamide, since some lipid oxidation products such as acrolein might react with asparagine.

Longer fermentation time of yeast doughs have been shown to produce bread with a lower acrylamide level since the yeast consumes the free asparagine. Sourdough as leavening agent is not as effective at consuming asparagine, probably due to competition from bacteria that impairs the yeast in the sourdough. If sourdough is to be used it is however better to use one based on oat or barley compared with rye or wheat flour since bread baked on oat or barley sourdoughs has been shown to have the lowest acrylamide content.

Acrylamide formation occurs in oat products, but the process is not as well studied as in other foodstuffs and more research is needed. This report aims therefore to list mitigation strategies to lower acrylamide formation in oat products.

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1 Introduction

Before 2002, acrylamide was mainly known for its use in polymer synthesis and in sealants. A sealant called Rhoca-Gil composed of acrylamide was used in the building of the Hallandsås tunnel in Sweden 1997 (Reimegård, 2002). Workers in the tunnel and fish and cattle in the surroundings showed clear symptoms of intoxication (Livsmedelsverket, 2019). Therefore, the workers were tested and high levels of acrylamide were found in their blood; but also in the control group in Malmö. Following studies found acrylamide in the whole population and not just in the workers of the Hallandsås tunnel or the animals in its surroundings (Magnusson, 2015; Reimegård, 2002). The reason was yet to be answered. In 2002 the Swedish Food Agency published a report which showed that acrylamide is produced during heating of starch-rich foods (Tareke et al., 2002). This was shocking since acrylamide is classified as a carcinogenic compound to humans (FoodDrinkEurope 2019). Since that, a plethora of research has been invested in acrylamide formation in foods. The foods of interest have often been based on wheat, rice and potato since these are some of the most important crops on our planet. The estimated production of wheat 2017 is around 772 million metric tonnes and for oats around 26 million metric tonnes (FAO, 2019). Therefore, oats have been less studied than wheat.

Some oat products have been found to have high levels of acrylamide (Capei et al., 2015). To solve this problem some mitigation strategies are needed. The European Union came out with legislation, (EU) 2017/2158 in 2017, which sets new and lower benchmark limits for acrylamide content in various food products than previous legislation. Since 11th April 2018 are food producers within EU strongly recommended to adapt suitable actions to make sure that their products do not exceed the new benchmark levels. There is however discussion within EU about changing these benchmark levels into strict levels that everyone must follow. The benchmark limit for oat-based breakfast cereals is 150 µg/kg and 300 µg/kg for wheat- and rye-based breakfast cereals. ((EU) 2017/2158, 2017) Rye and oats have higher acrylamide forming potential than wheat so one could argue that these benchmark limits should be changed. It would be more sensible to have limits at 150 µg/kg of acrylamide for wheat-based breakfast cereals and 300 µg/kg acrylamide for oat- and rye-based breakfast cereals.

Although, one Spanish survey of acrylamide levels in breakfast cereals found wheat-based cereals to have the highest acrylamide levels. High protein or fibre containing or puffed cereals had also among the highest of acrylamide levels. The tested products spanned from 62 to 802 µg/kg of acrylamide meaning that some products were far over the new EU benchmark limits (Rufian-Henares et al., 2006). Capei et al. (2015) measured acrylamide levels in oat-based breakfast cereals. Acrylamide levels were below limit of detection in some brands and as high as 450 µg/kg in other brands. This means that some of the brands had lower acrylamide levels than the benchmark limits set by EU meanwhile some exceeded them considerably. The study concluded that varying processing of the products between different brands rather than the raw material itself may be the reason for the differences between the brands (Capei et al., 2015).

A dietary risk assessment of acrylamide in Sweden 2003 found the mean consumption of acrylamide per day to be 35 µg. The increased cancer risk from a consumption of this magnitude stands in parallel to radon and background radiation. (Svensson et al., 2003) If one would eat a breakfast cereal with the acrylamide content equal to the new EU benchmark limit of oat-based breakfast cereals at 150 µg/kg, it would take around 230 g of breakfast cereals to reach an intake of 35 µg. This is more than one serving of breakfast cereals but since other products such as coffee, potato crisps and bread also contain acrylamide this limit is easily reached.

Acrylamide is not present in cereals out on the field but may be formed when they get heated during food preparation. The formation of acrylamide can take different pathways but the most involves asparagine and reducing sugars (Capuano and Fogliano, 2011). It is only the free asparagine that will react and it can react on its own although it won't produce as much acrylamide as if a reducing agent is present (Mottram et al., 2002). It has been shown in wheat and rye that free asparagine is the determining factor of acrylamide and it is thought to be the same in other cereals as well (Curtis et al., 2014).

Many papers have been released on the topic of acrylamide formation and some specifically on acrylamide formation in cereals and then mainly on wheat. These findings can be applied to oat products but if oats were not the one under scrutiny one must be careful before making assumptions.

This literature study aims to investigate acrylamide formation in oat products and to list approaches to limit its formation.

2 Literature findings

2.1 Acrylamide formation

In a study where bread made of wheat and wholegrain oat flour was baked, the acrylamide level in the crust was high. With sourdough as the leavening agent the pH dropped, and the acrylamide level was lowered by 10 %. It was also found that by covering the dough with a commercial asparaginase, the concentration of acrylamide in the crust dropped with 46 % and with no effect on the final quality (Ciesarova et al., 2014).

The amount of free amino acids in cereals may vary due to season, cultivars and harvesting time (Abdel-Aal and Hucl, 2002). One study found the free asparagine level in sifted wheat flour to be 87-89 (µg/g) with EZ-Faast method and 99 (µg/g) with the Biochrom method. Free asparagine levels in oat groats were found to be 431-443 µg/g and 525 µg/g with the same methods used (Mustafa et al., 2007). 25 wheat varieties in 2011-2012 and 59 wheat varieties in 2012-2013 grown in Great Britain have been studied. The grains were milled into wholemeal flour and the asparagine levels were tested with a slightly modified EZ Faast method. Free asparagine levels ranged from 93.5 µg/g to 1491 µg/g in different cultivars 2012-2013. 140 kg of nitrogen and 40 kg of sulphur per hectare were added to the fields. Ten of the varieties from 2012-2013 experiments had also been grown 2011-2012 with a rate of 130 kg of nitrogen and 40 kg of sulphur supplied per hectare. The range of free asparagine in 2011-2012 were narrow, ranging from 201 µg/g to 355 µg/g for the 25 tested varieties. Horatio, Croft, Myriad and five other tested varieties had low free asparagine levels in both 2011-2012 and 2012-2013's tests. This indicates, although environmental factors are important, that some cultivars have a lower free asparagine level. (Curtis et al., 2018)

Another study found the asparagine level to be 426 µg/g in wheat flour and 893,9 µg/g in oat flour (Žilić et al., 2017). This study aimed to compare different cereals in bread with each other with regards to potential acrylamide formation. Rye and oat cultivars had the highest amount of glucose, fructose and free asparagine. The study concluded that rye had the highest acrylamide forming potential and oats the second highest. The wheat varieties studied had the lowest potential. (Žilić et al., 2017) 26 cereal cultivars were grown in total and four of these were hull-less oat species. All cereals were grown outside of Belgrad during the harvesting season of 2015 and every analysis were duplicated for each cultivar. Sugar and free asparagine content of oat varieties are listed in *Table 1*. The total amount of sugar was around 1.7 % for all varieties while the asparagine content varied more. The lowest amount of asparagine was in JO-165 with 510.7 µg/g which is roughly 0.05 %. This means that there is roughly 34 times more sugar than asparagine in this flour. But the molecular weight also plays a role. Since the molecular weight of glucose and fructose (180 g/mol) is higher than the molecular weight of asparagine (132 g/mol), more sugar is needed weight wise to form acrylamide. However, sugars are still in excess, around 25 times more, after adjusting for the differences in molecular weights. These values show that the oat variety JO-165 had the lowest acrylamide forming potential due to the low amount of free asparagine. The report concluded that breeding and selection of varieties with a low amount of acrylamide precursors is one of the best mitigation strategies of acrylamide formation. How sugars are genetically connected with free asparagine production in cereals was also studied. Oat was the only studied cereal where there was no connection between free asparagine and sugar levels. (Žilić et al., 2017)

Table 1. Four oat varieties with sugar (%) and free asparagine amount (µg/g) tested. The cultivars were grown outside of Belgrade during 2015 and the analyses were done in duplicates. Galactose and maltose were found in other cereals but not in oats. “Total sugars” is the sum of all sugars. “Reducing sugars” is the total amount of reducing sugars.

Oat variety	**Glucose**	**Fructose**	**Sucrose**	**Total sugars**	**Reducing Sugars**	**Asparagine**
NS Sedef	0.54	0.35	0.85	1.74	0.89	1196.6
JO-50	0.61	0.37	0.90	1.88	0.99	887.8
JO-270	0.52	0.33	0.82	1.67	0.85	980.3
JO-165	0.50	0.33	0.83	1.66	0.83	510.7

Data were obtained from (Žilić et al., 2017)

How flour mixes of wheat, oat, soy, rye and maize flour and wheat bran affect acrylamide formation in breads have been studied. The AACC (1983) Method 10-10B were used to prepare the doughs. To maintain good baking properties, 70 % of the flour mixes were always wheat. Breads with all possible combinations of these flours were baked (wheat and one other flour, wheat and two other flours up to wheat and five other flours). The highest acrylamide levels were found in the control sample, baked on 100 % wheat flour and the bread baked on wheat and rye with 19.4 ± 0.6 μg/kg and 19.7 ± 1.1 μg/kg, respectively. Wheat and oat bread had a slightly lower concentration at 17.2 ± 0.7 μg/kg. In general, bread baked on flour mixes had less acrylamide compared with the control, spanning around 5 μg/kg of acrylamide. One exception was however the bread baked on wheat, oats, soy flour and wheat bran which had 19.4 ± 0.6 μg/kg of acrylamide. Most breads, except for the one just mentioned, with wheat bran in it had a low formation or none at all of acrylamide. However, no bread exceeded the EU benchmark limit of 50 μg/kg for wheat-based bread or 100 μg/kg for non-wheat-based bread (Serpen et al., 2012) It is counterintuitive that bread with wheat bran had a lower amount of acrylamide than wheat-based bread, since wheat bran has more asparagine compared to wheat endosperm and should therefore produce more acrylamide when heated. However, wheat bran binds more water than sifted wheat flour which might explain this reduction of acrylamide if the theory proposed by Rydberg et al. (2003), that lower water activity decreases acrylamide formation, is true. This theory has been tested. When oat bran concentrate with high fibre content and a high water binding capacity was added to a rye crisp bread it resulted in no acrylamide reduction (Mustafa et al., 2005). French fries have been studied and during the processing the water activity on the surface dropped, which enhanced acrylamide formation (Matthaus et al., 2004). Ciesarová et al. (2011) studied aspects affecting acrylamide formation in model systems based on potatoes and wheat. Water content were found to have a distinct impact on acrylamide formation and with the least acrylamide formed at 15-45 % moisture. Below 15 % moisture more acrylamide was formed which does not support Rydberg's theory. The fact that the least acrylamide was formed between 15-45 % moisture complicates the theory since it does not seem to be a linear connection between water activity and acrylamide formation. A possible reason might be the cooling of the system when water evaporates which lowers the temperature and therefore lowers the acrylamide formation. A similar theory for acrylamide reduction has been suggested by Capuano et al. (2010).

2.2 Precursors for acrylamide

Maillard reactions is the name of a series of reactions which contributes with flavour and aroma to cooked foods. It might however also lead to formation of acrylamide. Different types of reducing agents can interact in the *Maillard* reactions, leading to the formation of acrylamide, and sugars is one of them (Mottram et al., 2002). Free asparagine alone could under heating produce a negligible amount of acrylamide.

With the presence of a reducing agent the yield increased and with glucose present it was up to 80 times more acrylamide formed compared with asparagine alone (Zyzak et al., 2003). Further studies have therefore examined the sugar content of cereals. Sucrose in *Maillard* reactions, needs to undergo hydrolysis and form glucose and fructose. These sugars have reducing properties and can now react with asparagine to form acrylamide. The degree of hydrolysis of sucrose during cooking determines in the end how much acrylamide is formed (Gökmen et al., 2007). Which sugar that together with asparagine produces the most acrylamide depends on which state the sugar is in. In low moisture conditions the sugars will be in crystalline form and *Maillard* reactions will start to take place when the sugars react and form *Strecker* derivates as a first step of the *Maillard* reactions. This happens for fructose at 126 °C, glucose at 157 °C and galactose at 172 °C. Therefore, fructose has the highest molecular mobility and will at lower temperature come in contact with asparagine which enables acrylamide formation (Robert et al., 2004).

In systems with high moisture content may reagents move freely, and molecular mobility is less important. The decisive aspect for acrylamide formation under this condition is the chemical reactivity of the precursors. Thus, glucose and galactose show a higher reactivity than fructose in these kind of systems, since aldoses are more reactive than ketoses. (Robert et al., 2004)

Asparagine has been shown to be the determining factor for acrylamide in cereal products. Added sugar have therefore not shown any effect on acrylamide formation. This was evident when fructose was added to a rye crisp bread and no extra acrylamide was formed (Mustafa et al., 2005). However, by changing a reducing monosaccharide to sucrose in a cookie batter one can reduce the acrylamide formed by 50 % (Gökmen et al., 2007).

Free asparagine in cereals rarely exceeds 10 % of the total amount of free amino acids in the kernels, meanwhile it can be up to 30 % in potatoes. Although, 10 % can be exceeded under severe sulphur deficiency if nitrogen is present in extent. A situation like this in barley plants lead to that 50 % of the total amount of free amino acids were asparagine. (Halford et al., 2007) This have also been shown in wheat cultivated under sulphur depletion. The sulphur depleted wheat was compared with a control which received normal amounts of sulphur. The free asparagine level in the sulphur depleted crop were 30 times higher compared to the one with sulphur at normal levels. The kernels were then milled into flour and heated for 20 minutes at 160 °C. The acrylamide in the sulphur depleted sample were between 2600-5200 µg/kg and 600-900 µg/kg in the control. (Muttucumaru et al., 2006) The leading theory is that the plant uses asparagine as a nitrogen storage since it has a low C/N ratio and that asparagine is the substrate for only a few plant enzymes and may therefore be regarded as an unreactive compound. The asparagine accumulation is also induced by drought, pathogen attacks, high salt levels, lack of nutrients (sulphur, phosphorus, magnesium, potassium and zinc) and exposure to toxic metals such as cadmium and lead. (Lea et al., 2007).

The free asparagine is stored more in the bran of wheat and rye compared to the endosperm. Two wholemeal flour of wheat and one of rye had 510, 480 and 1070 μg/g DW free asparagine respectively. The same species' sifted flours had 140 and 170 μg/g for wheat and 530 and 680 μg/g DW for rye of free asparagine. The highest amounts were found in germ fractions where two wheat samples had 4880 and 4990 μg/g DW of asparagine. (Lea et al., 2007)

One study examined how different flours affected the formation of *Maillard* reaction intermediates as an indicative measure of the final formation of hydroxymethylfurfural (HMF) and furfural. Although acrylamide was not in focus, the results are still applicable since its formation involves the same reactions and compounds. Flour and water were mixed in sealed tubes and then heated at 150 °C for 30 minutes and the flours tested were oat, rye, wheat, spelt and teff. Methylglyoxal (MGO), glyoxal (GO) and 3-deoxyglucosone (3-DG) were measured in each sample since these are intermediates in the *Maillard* reactions. Glucose was also added to another set of samples to enhance the *Maillard* reactions. Wheat, spelt and rye without added glucose had the highest MGO and GO concentrations. Oats and teff had the lowest concentration of MGO and GO respectively. Glucose addition caused an increase in the formation of all three intermediates in all flours. With glucose, oats had as high concentration of MGO as the other flours except of spelt which had a slightly lower value. Wheat with glucose had also the highest GO amount and the most 3-DG was found in oats. Teff had by far the highest formation of HMF and furfural meanwhile wheat and oats had the lowest. The writers conclude that the increasing demand for gluten free alternatives makes the producers test new types of flours. This can be a food safety risk since alternative flours are not as well studied as traditional wheat flour. For the same reason might this be a new useful tool for reducing acrylamide formation, but more research is needed. (Mesías and Morales, 2017)

2.3 The effect of pH and antioxidants on acrylamide formation

Changes in pH affect the extent of acrylamide formation. One study mixed potatoes and then heated them at 180 °C for 25 minutes before measuring acrylamide content. A maximum of 800 μg/kg acrylamide was found at pH 8. Acrylamide content decreased alongside with a decrease in pH down to to 200 μg/kg acrylamide at pH 3, which was the lowest tested pH. (Rydberg et al., 2003)

Ou et al. (2010) tested if antioxidants can inhibit acrylamide formation in aqueous asparagine and glucose models or directly deplete it when pure acrylamide were mixed with water. Tested antioxidants were: tert-butyl hydroquinone, butylated hydroxy anisole, butylated hydroxytoluene, ferulic acid, epigallocatechin gallate and vitamin C and their respective oxidised forms. The antioxidants could not degrade acrylamide or stop its formation, but their oxidised forms could directly destroy acrylamide and asparagine and thereby limit its formation.

Contrary data have however been reported. A study where ascorbic acid was added to a potato sample, with a final concentration of 1.51 mmol/kg, which was then oven-heated found that the acrylamide concentration increased (Rydberg et al., 2003). The same study also found that at extreme concentrations of ascorbic acid, 123 mmol/kg, the acrylamide formation dropped with 46 %. This could be explained with the drop in pH as well as the inhibition of water to react in acrylamide formation since ascorbate binds to it. They concluded that not just ascorbic acid but also other compounds that bind water will decrease the acrylamide formation since water prevents pyrolysis reactions (Rydberg et al., 2003).

2.4 The effect of lipid oxidation on acrylamide formation

Oxidation products of rancid frying oils can react with asparagine and form acrylamide (Ou et al., 2010). Peroxide value measures primary oxidation of oils meanwhile carbonyl value measures the secondary oxidation into aldehydes, ketones and other carbonyl compounds. Heating of oil increases carbonyl value and forms a compound called acrolein. Acrolein can together with asparagine form acrylamide. Presence of antioxidants in frying oils might inhibit oil oxidation and thus prevent this process and lower the amount of acrylamide formed (Ou et al., 2010) Fat content of oats varies with cultivars and have been found to span between 4.2 to 12.4 g per 100 g. Around 80 % of the fatty acids are unsaturated which paves the way for oxidation. (Sterna et al., 2016) (Zhou et al., 1999). But lipid oxidation's role on acrylamide formation is also affected by other factors and one of them is water content. This have been shown in models systems consisting of 35 % oil, 4 or 16 % water, reducing sugars and added thickeners to achieve a dough-like structure. 1 g of the dough was heated at 180 °C for 30 minutes and acrylamide content were measured. Oxidation products were showed to be able to convert asparagine into acrylamide. In the dryer models, the final acrylamide content was found to be higher than in the one with 16 % water. A proposed theory is that higher initial water content cools the inside of the dough during baking and lower temperature yields less acrylamide. With reducing sugars present the oxidation products had a lower impact on the final acrylamide content since the reducing sugars reacted first. Lastly, it was concluded that addition of catechins (an antioxidant) lowered the acrylamide formation the most in low moisture models with relatively low carbohydrate concentrations and where the majority of the acrylamide is formed from lipid oxidation products (Capuano et al., 2010).

If the oil used in baking have been thermal oxidised it leads to more acrylamide. Arribas-Lorenzo et al. (2009) studied the effect of thermal oxidised sunflower oil in baking of cookies. After 14 minutes, the thermal oxidised oil produced no higher amount of acrylamide but after 16 minutes, only two minutes later, the cookies with thermal oxidised oil had 59 % higher acrylamide content compared with the control. This sudden increase points to that lipid oxidation products affect acrylamide formation at certain conditions; since inner moisture decreases and temperature increases over baking time. Oils with various amounts of phenolic compounds also affected the acrylamide content. Phenolic compounds have antioxidant activity and could thus lower acrylamide formation, although as previously mentioned some studies have found that antioxidants increased acrylamide formation (Rydberg et al., 2003). Oil with higher amount of phenolic compounds yielded lower amount of acrylamide, and this effect became clearer as baking time increased (Arribas-Lorenzo et al., 2009). It is although unclear if oat’s fat content affect acrylamide formation.

2.5 The effect of salts on acrylamide formation

Most breakfast cereals have a heating step in the production. During this step acrylamide can form. It has however been discovered that the presence of sodium chloride may reduce the acrylamide formation significantly ($p < 0.05$). To add more salt to cereal products may therefore decrease the overall acrylamide consumption but this will naturally increase the salt intake (Moreau et al., 2009). WHO recommends that one should not consume more than 5 g salt per day since a high salt intake correlates with increased risk of cardiovascular diseases (World Health Organization, 2007). Average daily salt intake in the western world is 9-12 g per person and bread and breakfast cereals is the reason for a third of this amount (Moreau et al., 2009). The study concluded that salt interacts with *Maillard* reactions and colour formation, which usually goes hand in hand. This highlights the problematic approach to estimate acrylamide formation based on colour observations since presence of salt may change the browning of the product. Colour is also an important quality parameter for the customers and addition of salt makes the product darker. (Moreau et al., 2009).

The same study also examined if salt interacts with *Strecker*-degradation, which is an early step in the *Maillard* reactions, by analysing how much of some volatile aldehydes, known as *Strecker* aldehydes, were formed in a model system. These volatiles have been claimed to be important for the aroma of fried foods. Salt did not affect the amount of aldehydes formed and thus the report concluded that salt probably affects the later steps of the *Maillard* reaction since lesser acrylamide was formed (Moreau et al., 2009).

Other salts than sodium chloride affect the acrylamide formation. When a cereal model system was heated for 9 minutes at 190 °C the presence of calcium chloride could lower the acrylamide formation with 90 %. Potassium dihydrogen phosphate and sodium acid pyrophosphate lowered the levels of acrylamide with 75 % while ammonium hydrogencarbonate increased it. The increase of acrylamide caused by ammonium hydrogencarbonate could be due to the fact that it increases the pH which favours acrylamide formation. Calcium chloride is potent for acrylamide lowering, however it has affected the final taste of products and in a taste trial of sweet biscuits it inhibited the rise of the biscuits and rendered them inedible (Kukurová et al., 2009)

3 Solutions and mitigation strategies to lower acrylamide formation in oat products

Wheat, rye, barley and oat have been studied as mediums for sourdoughs with a pure line of *Lactobacillus plantarum*. Various aspects of a rye and wheat bread (60/40; rye/wheat flour; w/w) baked with these sourdoughs were measured and it was found that acrylamide levels in breads baked on barley and oat sourdoughs were below 11 μg/kg but as high as 80 μg/kg when baked with wheat sourdoughs and around 50 μg/kg with rye sourdoughs (Bartkiene et al., 2017). The EU benchmark levels for acrylamide in wheat bread is 50 μg/kg and 100 μg/kg for non-wheat breads ((EU) 2017/2158, 2017). The test values can be regarded as quite high since most of the flour is wheat. Bartkiene et al (2017) also found that bread baked on wheat and rye sourdoughs produced more of the D-isomer of lactic acid which another study has shown to be detrimental to human health (Jin et al., 2016). Breads baked with oat sourdoughs had a 12:1 L/D-ratio of lactic acid meanwhile rye and wheat sourdough breads had a 10:1 L/D-ratio. It is notable that sourdoughs affect more parameters of a bread such as moisture content, bread shape and overall acceptance. At a low percentage at 5 % of oat sourdough starter, the best shape, i.e. highest bread slice, was obtained. The breads baked with rye sourdough starters had the highest acceptance (Bartkiene et al., 2017).

The fermentation time of yeast doughs also affects the final acrylamide content. A wheat flour and wheat bran bread with a total rising time of 30 minutes have been compared with a bread constituting of the same flour but with a fermentation time of 360 minutes. The bread with a longer fermentation time had 87 % lower acrylamide compared to the other bread. When sourdough was used it showed a lower decrease of acrylamide compared to when yeast was used. A reason for this might be that the bacteria interfere with the yeast in the sourdough and inhibiting it from consuming the free asparagine in the dough. (Fredriksson et al., 2004) Several other studies have come to the same results, that yeast fermentation decreases the asparagine content in the dough and thus decreases the acrylamide content in the baked bread (Wang et al., 2017, Sadd et al., 2008, Esfahani et al., 2017) Acrylow^{TM} is a yeast, developed especially to consume asparagine. Depending on product and application have Acrylow^{TM} been shown to reduce the final acrylamide content with 50-95 % (Renaissance Ingredients, 2019).This yeast is quite new and more external testing is needed to say if it is as good as the company claims; although the results so far seem promising.

Flatbreads based on several alternative flours have been made to determine if other flours than traditional wheat might produce flatbreads with lower acrylamide concentration. Whole oat flour along with some other flours had very low acrylamide concentrations (<10 µg/kg). The wheat-based commercial counterparts had mostly higher acrylamide concentrations and especially matzo bread which had between 101 and 2070 µg/kg. (Crawford et al., 2019) Comparatively, the benchmark acrylamide limit for non-wheat-based bread is 100 µg/kg; meanwhile the limit is 50 µg/kg for wheat-based bread ((EU) 2017/2158, 2017). Changing from wheat to oat flour might serve as a mitigation strategy since it has been shown to lower acrylamide formation (Crawford et al., 2019) This decrease contradicts the fact that oat has higher acrylamide forming potential than wheat. Nevertheless, changing from wheat to oat flour will make the product to end up in another benchmark category with a higher acrylamide limit.

By adding alanine, glycine, lysine, glutamine and glutamate to potato samples which underwent heating at 180 °C a decrease in acrylamide formation were achieved. The decrease ranged from 14 % when 35 mmol/kg alanine was added up to 92 % when 140 mmol/kg glutamine was added. (Rydberg et al., 2003)

Asparaginase is naturally present in plants and plays a central role in the energy production by dividing asparagine into aspartic acid and ammonium. The same principle can be applied to foods. By reducing the precursors of acrylamide one can inhibit its formation. L-asparagine is the most important precursor and by adding L-asparaginase extracted from various bacteria and fungi to starch-rich products it has been showed to lower the acrylamide formation. 86 % and 94 % reduction of acrylamide formation have been achieved, compared to controls, in wheat bread and potato chips treated with L-asparaginase during the production (Meghavarnam and Janakiraman, 2018). Asparaginases have been extracted from *Escherichia coli, Bacillus sp., Erwinia carotovora, Candida utilis, Corynebacterium glutamicum, Pseudomonas stutzeri and Enterobacter aerogenes* and applied to foods with various success of preventing acrylamide formation. Most asparaginases are active around pH 5-7 and at 30-70 °C and will therefore be inactivated during cooking. Since glutamine has a similar structure to asparagine might some asparaginases also degrade it into glutamic acid and ammonium. The survey of Xu et al. (2016) compiles research where asparaginases have been used to lower acrylamide in foods. To better know how much of asparaginase to use, one should know the amount of asparagine in the raw product and control pH and temperature. The conclusion made is that asparaginase may be the key to solve the acrylamide problem but the industrial application of it is limited since it has yet too adverse effects on final quality. (Xu et al., 2016)

4 Discussion and conclusions

Acrylamide formation in oat products is not as well known as in wheat and potato products. This is a risk since studies have found oats to have the second highest acrylamide forming potential of the four kinds of cereals. Fried, roasted, toasted and extruded products cannot be achieved without some heating step. The demand for these types of products will probably not decrease over time. Therefore, it is important to control pH, moisture content, heating time and the amount of acrylamide precursors in the start product. The latter can be done over time with breeding towards crops with low levels of free asparagine and by controlling the fertilisation to minimise the formation. More free asparagine is formed when there is a lack of sulphur and at the same time an overflow of nitrogen. Avoidance of drought, to high salt concentrations, toxic metals and poor nutrition on the crops will also reduce the amount of free asparagine being formed.

Different sugars react at various rates to form acrylamide. Fructose and glucose form more acrylamide than sucrose in general. This is however of little significance for cereal products since asparagine have been shown to be the determining factor. If the heating step is short however it might be relevant since fructose and glucose reacts faster than other sugars. If the heating is just above 120 °C mainly fructose reacts. Which sugar that is used may therefore affect the final acrylamide content. By preventing hydrolysis of sucrose, one will also lower the acrylamide formation since sucrose is not a reducing sugar but its constituting sugar units, glucose and fructose, are.

Since *Maillard* reactions do not occur below 100 °C, within the time for cooking food, boiling is regarded and has been proven to be a safe way of preparing starch rich foods. Although, boiling in a pressure cooker could maybe lead to acrylamide formation since it can reach a temperature of 120 °C which is when fructose starts to react with asparagine and form acrylamide. This is only a theory however since acrylamide is not usually produced when a lot of water is present.

Addition of inorganic salts may be a mitigation strategy of acrylamide and a highly potent salt is calcium chloride. This salt affects the final product quality but may be useful if applied in the right way.

Addition of asparaginases can also serve as a tool for lowering the amount of acrylamide formed but it is still on experimental level for now and not used widely in the industry due to off-flavours being produced. Application of yeast which produces asparaginases and lowers acrylamide formation seems more promising. This mitigation strategy works well in products with a fermentation step such as coffee, bread and biscuits.

The degree of oil oxidation in deep fried products has been shown to affect the extent of acrylamide formed. Oat products are rarely deep fried but oats on the other hand can contain a considerable amount of fat. This fat is also mostly unsaturated and could oxidise. Oats however, contain polyphenols which have antioxidant activity which have been shown to inhibit acrylamide formation. It is therefore hard to say if lipid oxidation plays a major role in acrylamide formation in oat products, but it is at least worth a thought and maybe continued research.

Broadly speaking, there are two categories affecting acrylamide formation, the raw materials and the processing. The processing step often includes *Maillard* reactions which may produce acrylamide. By applying less heat (time and temperature) the *Maillard* reactions will decrease. This will however also affect the formation of flavour and aroma compounds which it also contributes with. One can therefore only modify the processing to a certain extent without affecting the final quality of the product. Changing the raw materials might be costly but could lower the acrylamide formation more effectively. Appliance of salts, asparagine-consuming yeasts and breeding towards cereal crops with lower free asparagine and higher antioxidant levels might thus be the way forward in solving the acrylamide problem.

Conclusively, more research about acrylamide formation in oat products is needed to know to which degree oat products contribute to the total acrylamide exposure of humans and what remedial actions that should be applied.

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Acknowledgements

Thanks to Roger Andersson, professor at Department of Molecular Sciences at SLU, for providing his expertise in this field and setting me up with CG Pettersson at Lantmännen who came up with the idea for this work. I am thankful for the idea and would like to thank CG Pettersson for letting me investigate further in it. I would also like to thank Xue Zhao, PhD student at Department of Molecular Sciences at SLU, for insightful comments about my writing and continuing help with references during the work.