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Storage of potatoes at low temperature should be avoided to prevent increased acrylamide formation during frying or roasting

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Introduction

Acrylamide is of concern as a probable carcinogen in some strongly heated foods (1, 2). It may be present at concentrations exceeding 1000 µg/kg in foods consumed in large quantities (e.g. (3)), such as roasted or fried potato, i.e. exposure is high compared to that of other suspected carcinogens. As for most substances classified as carcinogens, there is no ultimate proof for carcinogenicity in humans, but application of the precautionary principle requires that reasonably feasible measures should be taken to reduce the exposure of consumers.

Acrylamide is formed by thermal degradation of the free amino acid asparagine supported by fructose and glucose (4–8). The reaction takes place at temperatures above some 100°C and presupposes a fairly dry matrix, i.e. typically occurs during baking, frying or roasting. Particularly high asparagine concentrations are found in potato (typically 2–4 g/kg referring to fresh weight, i.e. some 10–20 g/kg per dry mass (9)); wheat flour only contains around 0.15–0.4 g/kg asparagine (10 samples; unpublished data).

As shown in ref. (10), for most consumers, the fried, roasted or baked potato products are the predominant sources of acrylamide: easily 95 % of the total exposure for a person eating three times roast potatoes ("Rösti") per week or some 85 % for somebody three times eating French fries. For this reason, Rösti, oven-roasted

potatoes, French fries and potato chips should be given top priority in searching for ways to reduce acrylamide formation.

Acrylamide contents in French fries, roast potatoes and similar products can be reduced by a factor of about five to ten by use of better potatoes and adjustment of preparation conditions. *Gertz and Klostermann* (11) and the official food control of Stuttgart (Chemisches und Veterinäruntersuchungsamt, (12)) advocated lower temperatures for preparing French fries. In collaboration with the School of Hotel Management Belvoirpark, Zurich, the steps for preparing French fries of optimum culinary quality and minimum acrylamide content were optimized, starting from the selection of appropriate raw potatoes (10). This enabled to reliably achieve French fries with 40–70 µg/kg of acrylamide (compared to the currently normal 200–800 µg/kg). On-going work on roast potatoes (Rösti) shows a similar potential of reducing acrylamide contents.

Acrylamide formation in a potato product strongly depends on the quality of the raw potato, particularly on its content in reducing sugars. The tendency of a potato to produce acrylamide upon heat treatment was measured by the “potential of acrylamide formation” (13), a standardized and reproducible heat treatment at 120°C for 40 min. This potential of acrylamide formation served to compare potatoes. It does not enable to calculate acrylamide concentrations in a specific food, but was shown to be proportional to the acrylamide concentrations in potato products prepared under constant conditions. *Amrein et al.* (9) showed that it is strongly correlated with the content of reducing sugars and asparagine. Since asparagine varies little, the far broader variation of the reducing sugars essentially determines acrylamide formation. With this, acrylamide formation is linked to the broad knowledge available about reducing sugars in potato. Reducing sugars are key substances for the quality of potato products like chips and French fries (14).

The potentials of acrylamide formation and the contents of reducing sugars in Swiss potato cultivars varied by a factor of about 30 (9), i.e. a given heat treatment causes some potatoes to form up to 30 times more acrylamide than others. The cultivars with the lowest contents in reducing sugars, e.g. *Erntestolz*, *Panda* and *Agria*, are those commonly used for the preparation of potato chips and French fries. Differences in farming, such as fertilization, seemed to be of minor importance (8, 9).

In practice, the acrylamide content of potato product is not really proportional to the potential of acrylamide formation. As the latter is linked with the Maillard reaction, it occurs parallel to browning. Roasting or frying potatoes rich in reducing sugars results in rapid browning, i.e. the cook tends to stop earlier than for potatoes with low sugar contents. Potato chips are an exception to this, since they are fried until the water content is below a given limit. French fries or Rösti prepared from potatoes with a high sugar content are either crispy and dark (and high in acrylamide content), or, if excessive browning is avoided, remain soft – they become dark more rapidly than crispy.

For potatoes, conditions of storage are even more important than the cultivar: when cooled below 8–10 °C, potatoes liberate reducing sugars, presumably to protect themselves against frost. This is a well known phenomenon also termed “low temperature sweetening” (15, 16). Near the freezing point, potatoes get so sweet that they are no longer considered edible. Since a temperature of 4 °C does not cause disturbing sweetening yet, this is the standard condition to suppress sprouting during long term storage. For potatoes of the cultivar Erntestolz, 15 days of storage at 4 °C increased the potential of acrylamide by a factor of 28 (13). The sum of the reducing sugars referring to fresh weight increased from 80 to 2250 mg/kg (8), i.e. also by a factor of 28.

This paper intends to draw the attention of the potato experts to the importance of storage conditions for acrylamide formation, providing results on the increase of the potential of acrylamide formation linked to reducing sugars. It collects data accumulated during July 2002 to March 2003, usually performed in parallel to experiments concerning other subjects. More thorough experimentation, properly taking into account the complexity of the matter, will be needed to conclude on the measures to be taken.

Experimental

Experiments on cooling potatoes were performed in a cooling room adjusted and regularly checked to be at 4 ± 1 °C. Storage in a cellar of the laboratory was at temperatures varying between 10 and 15 °C, the potatoes being covered up against light.

Potentials of acrylamide formation were determined as described in ref. (13) and refer to fresh weight. Of a given sample, five to ten tubers were halved. One half of each was grated. 20 g of the homogenated grated material was spread on a grid and heated in a GC oven to 120 °C for 40 min. Then the dry matter was weighed and acrylamide determined according to ref. (17). Water was added and the potato swollen at 70 °C. The sample was extracted using 1-propanol, the propanol/water mixture evaporated, the acrylamide dissolved from the residue into acetonitrile and this extract defatted with hexane. Analysis involved GC-MS with chemical ionization (single determinations). The relative standard deviations of determining potentials of acrylamide formation were 5–6 % (13).

Reducing sugars were determined enzymatically using the test kit from Scil Diagnostics (Martinsried, Germany) (8). Briefly 20 g of grated potato was blended with 60 g distilled water. 5 ml of solutions Carrez I (150 g/l potassium hexacyanoferrate(II) trihydrate; Merck, Darmstadt, Germany) and Carrez II (300 g/l zinc sulfate heptahydrate; Fluka, Buchs, Switzerland) were added. The mixture was thoroughly shaken, the pH adjusted to 7.0–7.5 with a few drops of potassium hydroxide solution (4 mol/l; Fluka), foam broken by addition of 50 µl of 1-octanol (Fluka) and the volume adjusted to 250 ml with distilled water. Filtered samples (Schleicher&Schuell) were subjected to enzymatic analysis as described by the producer.

Results

Storage above about 10 °C

In ref. (8), first results were reported for potatoes of the cultivar Charlotte from the garden of one of the authors. The potatoes were harvested on July 4, 2002, and had a 120 °C potential of acrylamide formation of 150 µg/kg. Stored in the natural cellar of the house at temperatures of 11–16 °C, the potential remained low (figure 1). In March 2003, the potatoes started substantial sprouting and had lost much water. Actually most of the increase of the potential observed in March (260 µg/kg) can be explained by the increase in dry weight. The sum of the reducing sugars initially fell from 0.3 g/kg to 0.125 g/kg, remained at this level for a long time and increased to 0.53 g/kg in March. This behavior is in agreement with data shown by Burton (18).

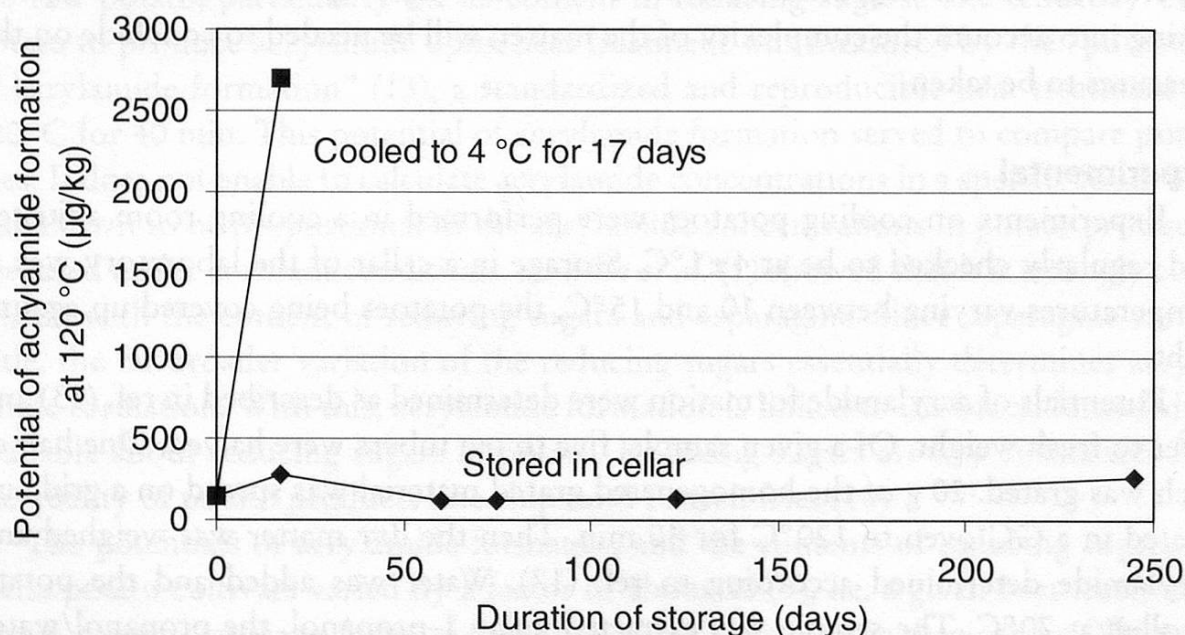


Figure 1 **Potential of acrylamide formation in potatoes of the cultivar Charlotte dug on July 4, 2002 and stored at 11–16 °C in a private cellar up to March 10, 2003**

In July 2002, a sample of these potatoes was stored at 4 °C for 17 days. The potential of acrylamide formation increased to around 2700 µg/kg and the content of reducing sugars to 6.4 g/kg fresh weight (i.e. by factors of 18 and 21, respectively).

Figure 2 shows the contents of reducing sugars for potatoes of cultivars typically used by the chips industry, stored at 10–12 °C from the beginning of October 2002 to the end of March 2003 in the facilities of Zweifel AG. Every data point represents two samples harvested in two different areas and stored up to the end of September at different places. Contents clearly decreased for a majority of the cultivars. The

Lady Rosetta is an exception: the sugars increased in March, probably related to the strong sprouting (Lady Rosetta is not suitable for long term storage). For all others, the summed concentrations of glucose and fructose were 300–500 mg/kg dry matter, i.e. hardly 100 mg/kg fresh weight, which is low indeed.

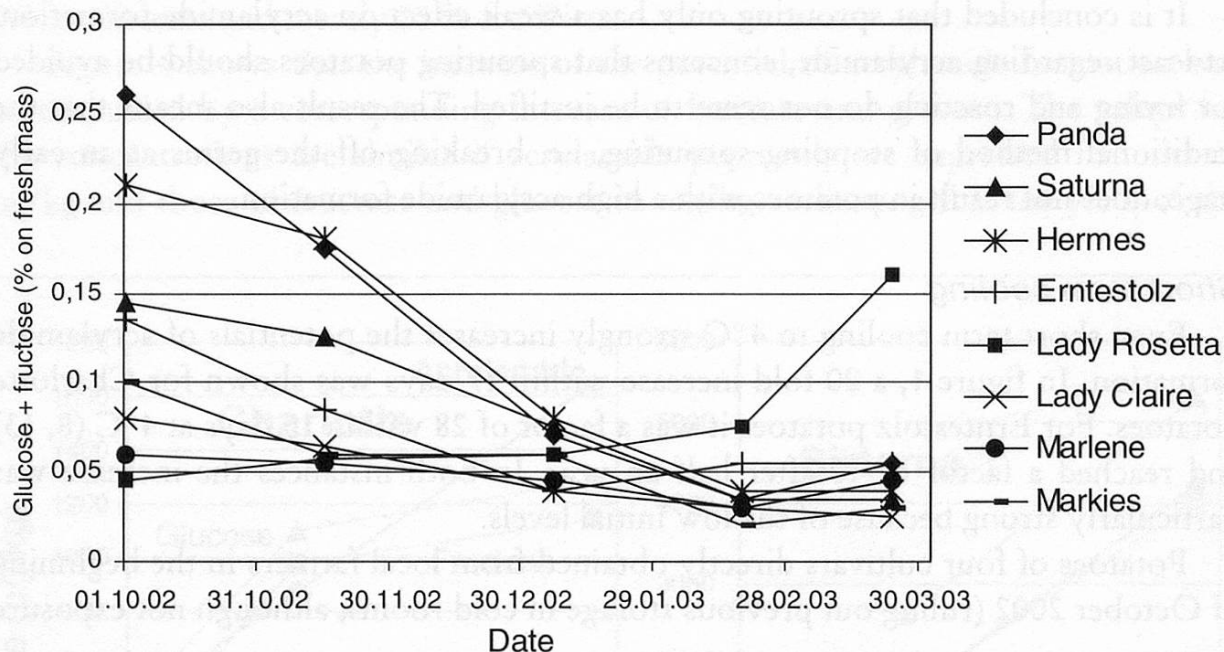


Figure 2 **Contents of reducing sugars for potatoes kept at 10–12°C from October to March**

Also among the potatoes stored in the cellar of the laboratory (cultivars Agria, Bintje, Urgenta, Ditta), none showed a clear increase in contents of reducing sugars or potentials of acrylamide formation up to January–March, 2003. It is concluded that storage at temperatures of at least 10°C at least up to March does not increase acrylamide formation.

Sprouting

The hypothesis could not be verified that sprouting strongly mobilizes reducing sugars and, hence, increases the potential of acrylamide formation correspondingly. Several experiments with weak sprouting (up to 5 cm) did not show a significant increase. Also the potatoes of the cultivars Hermes, Lady Claire and Saturna of figure 2 started sprouting without showing an increased sugar content.

To enhance a potential effect, an extreme case was investigated. Potatoes of the cultivar Urgenta were followed from their purchase at the end of August 2002 to March 2003. They were kept in the laboratory up to November 2002, when the first signs of sprouting became visible. The 120°C potential for acrylamide formation increased marginally from 340 to 400 µg/kg (probably explained by loss of water).

Then these potatoes were kept in the cellar at about 12°C. In the beginning of March, they sprouted some 25 cm high and the tubers were strongly shrunk. The potential of acrylamide formation raised to 900 µg/kg, the reducing sugars from 0.8 to 2.1 g/kg. Almost half of this increase was due to loss of water. The strongly sprouted Lady Rosetta (figure 2) showed a similar effect. In both instances, potatoes would no longer been considered edible.

It is concluded that sprouting only has a weak effect on acrylamide formation. At least regarding acrylamide, concerns that sprouting potatoes should be avoided for frying and roasting do not seem to be justified. The result also means that the traditional method of stopping sprouting, i.e. breaking off the germs at an early stage, does not result in potatoes with a high acrylamide formation.

Short term cooling

Even short term cooling to 4°C strongly increases the potentials of acrylamide formation. In figure 1, a 20 fold increase within 17 days was shown for Charlotte potatoes. For Erntestolz potatoes it was a factor of 28 within 15 days at 4°C (8, 13) and reached a factor of 40 after half an year. In both instances the increase was particularly strong because of the low initial levels.

Potatoes of four cultivars directly obtained from local farmers in the beginning of October 2002 (ruling out previous storage in cold rooms, although not exposure

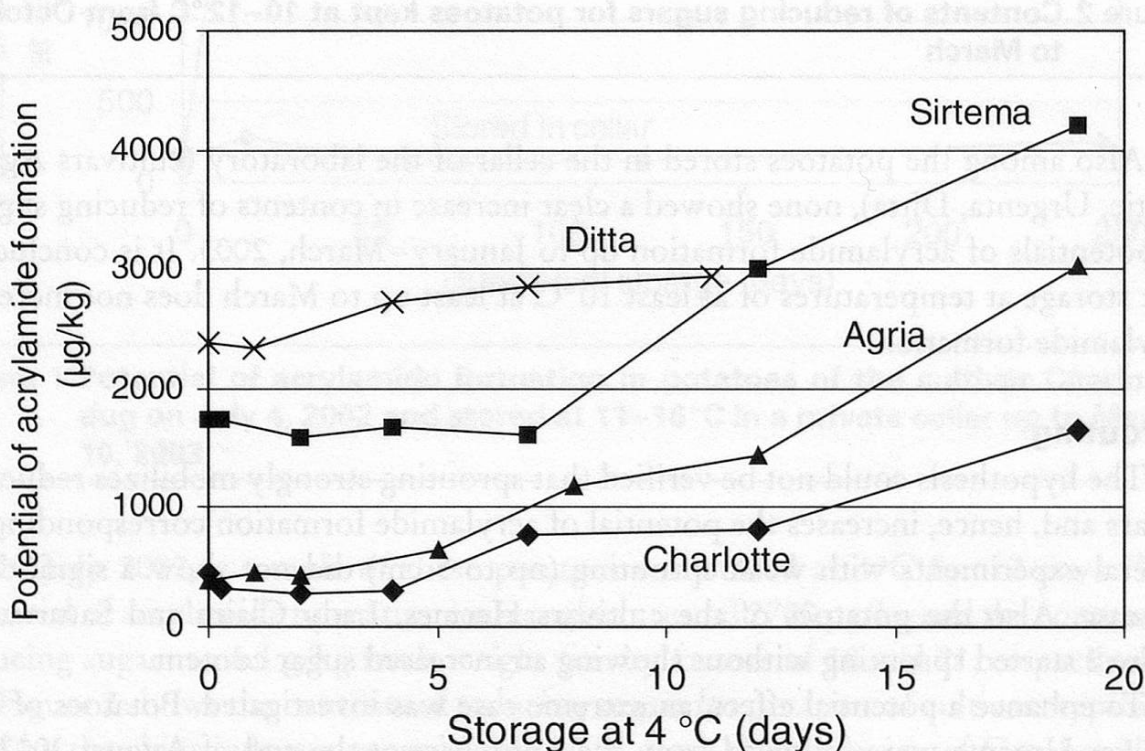


Figure 3 Potential of acrylamide formation in potatoes transferred to a 4°C room; first 19 days (for longer periods, see figure 6)

to cool air) were placed in the 4°C room on October 10, 2002. Samples were analyzed immediately after bringing them to the warm laboratory. Figure 3 shows results of the first 19 days. There seems to be a lag phase of around 5 days until the potentials of acrylamide formation started to rise. After 19 days, they had increased by a factor of 3.8 for the cultivar Charlotte and by a factor of 7.8 for Agria. For the cultivars with a high potential to begin with, the increase was more modest: a factor 2.4 for Sirtema and merely 23 % for Ditta.

Figure 4 shows that the increase of the potential of acrylamide formation was accompanied by a corresponding increase of fructose and glucose. The values for the concentrations are similar if acrylamide potentials are expressed in units of µg/kg and those of fructose and glucose in mg/kg, indicating that the yield of the

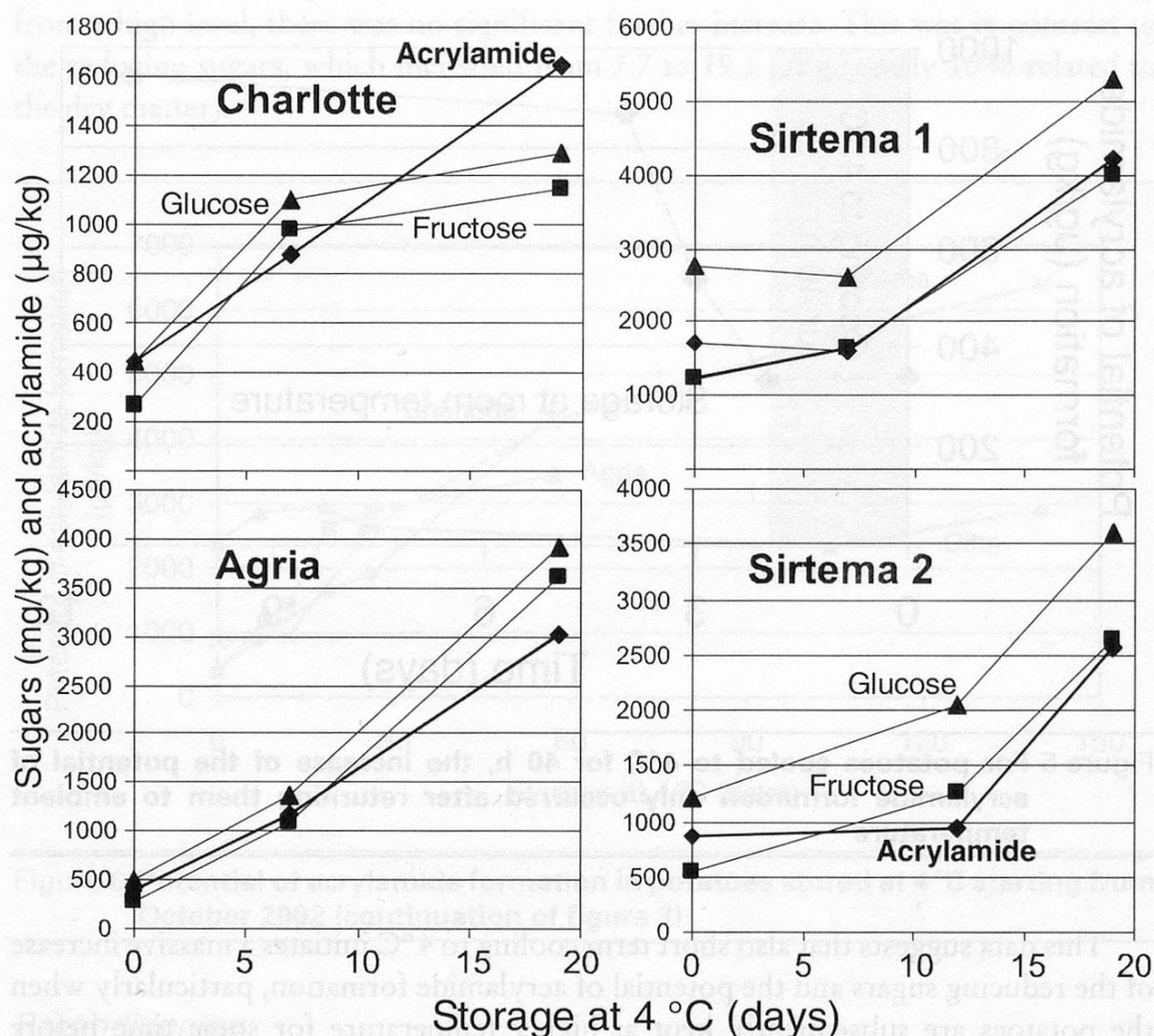


Figure 4 Increase of the potential of acrylamide formation (bold lines) and of the reducing sugars during the first 19 days of storage at 4°C; concentrations for fructose and glucose in mg/kg, those of acrylamide potentials in µg/kg

reaction is in the order of 0.1 %. The difference between the two samples of Sirtema was almost as large as between the cultivars. The samples of Sirtema potatoes were from different origins.

As suggested by figure 3, the increase of the reducing sugars and the potential of acrylamide formation occur with a time lag. This may also mean that the increase only occurs after the cooling period, as shown in figure 5. In September 2002, a sample of Urgenta potato was cooled to 4 °C for 40 h. The potential of acrylamide formation did not increase during this time, but only while the potatoes were at ambient temperatures again. The increase during the following two days was faster than in the cooling room (figure 3), probably because of higher enzyme activity at ambient temperature.

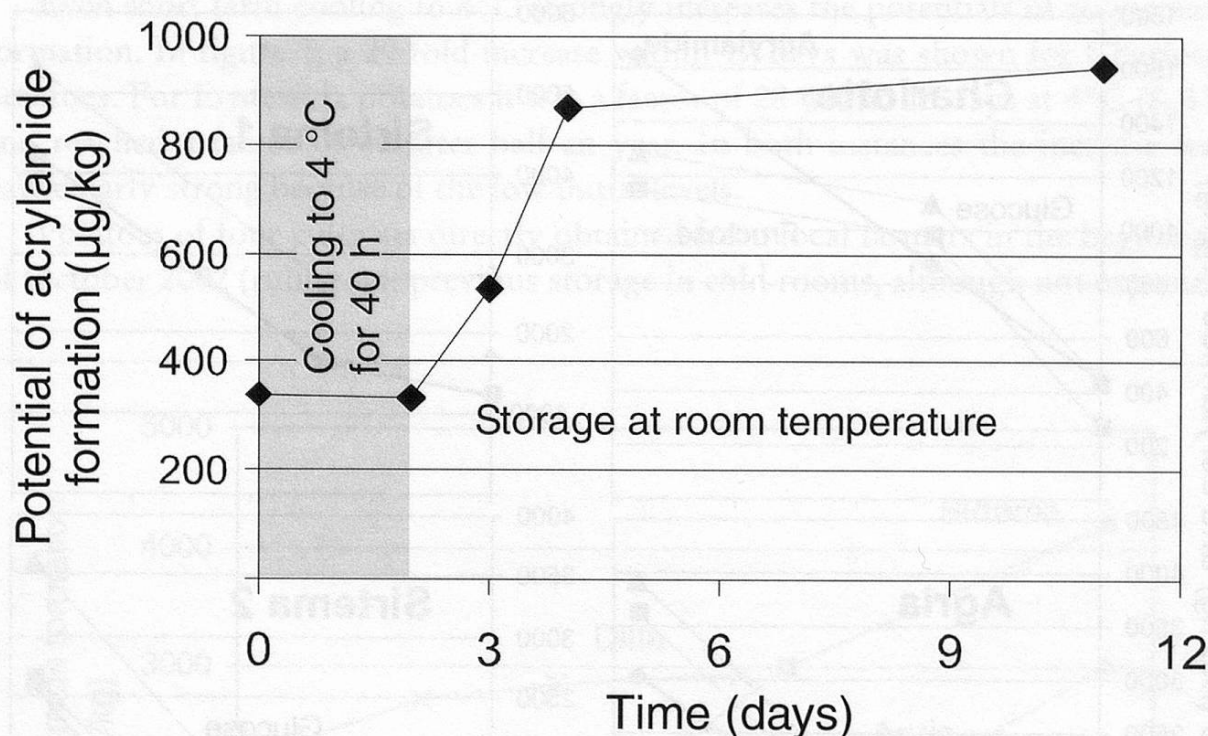


Figure 5 For potatoes cooled to 4 °C for 40 h, the increase of the potential of acrylamide formation only occurred after returning them to ambient temperature

This data suggests that also short term cooling to 4 °C initiates a massive increase of the reducing sugars and the potential of acrylamide formation, particularly when the potatoes are subsequently kept at higher temperature for some time before being used. Such cooling occurs in cooling rooms of distributors or restaurants as well as in the refrigerator of private homes. Short term cooling may also occur on the fields before or after harvesting or in the barn of the farmer before delivery to temperature-controlled storage.

Cooling of partially prepared potatoes, e.g. peeled and cut, for up to about two days, as frequently applied in restaurants, does not increase acrylamide formation provided the potato is then heated without remaining at ambient temperature for an extended duration (which is anyway avoided for reasons of hygiene).

Long term cooling

The standard temperature for long term storage delaying sprouting of potatoes is around 4°C. The effect of cooling in the cooling room of the laboratory is shown in figure 6 for a continuation of the experiment, the first 19 days of which were shown in figure 3. Starting in October, 60 days at 4°C increased the potential of acrylamide formation by a factor of 10 for Charlotte and 8.8 for Agria. Sirtema (different lot than figure 3) started from a relatively high level and further increased it by a factor of 7.3 up to March 2003. Ditta showed a particular behavior: starting from a high level, there was no significant further increase. This was in contrast to the reducing sugars, which increased from 7.7 to 19.1 g/kg (nearly 10% related to the dry matter).

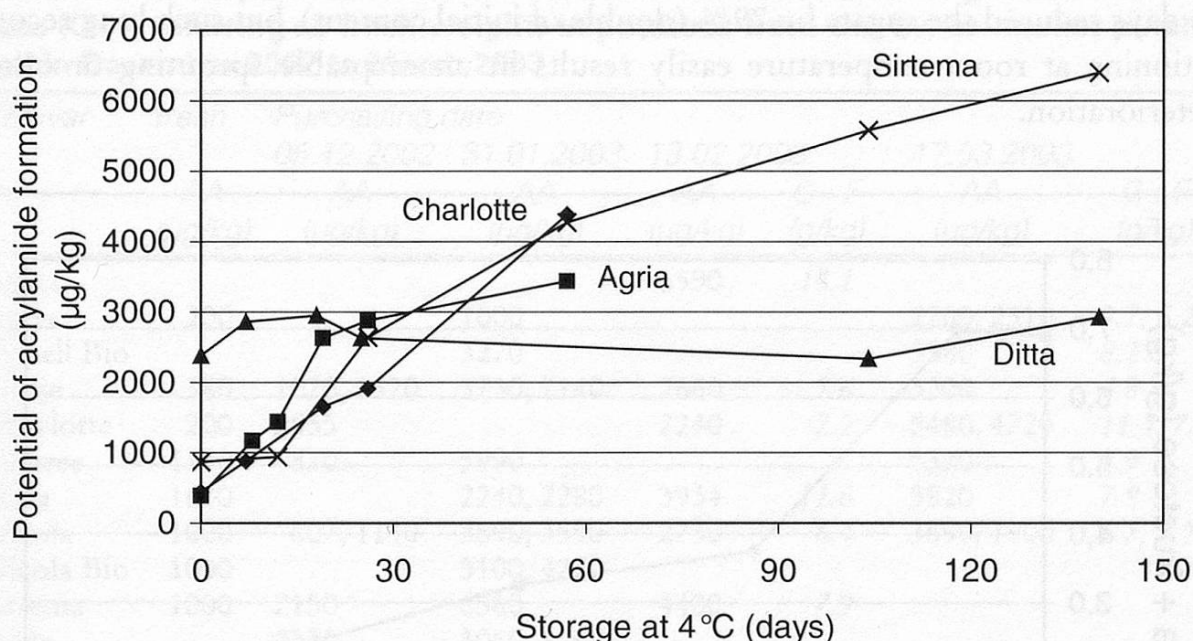


Figure 6 Potential of acrylamide formation in potatoes stored at 4°C starting from October 2002 (continuation of figure 3)

Reconditioning

When potatoes stored at low temperatures are reconditioned at higher temperature (12–25°C), the content of reducing sugars decreases again. The effect of short term (40 h) cooling on Urgenta potatoes followed by reconditioning at ambient temperature for three months is shown in table 1: the potential increased from

340 to 900 µg/kg and decreased again to 450 µg/kg, which is marginally significantly above the 400 µg/kg measured for the sample not submitted to cooling.

Table 1
120°C potentials of acrylamide formation for Urgenta potatoes stored as indicated

Potato	Sprouting	Acrylamide (µg/kg)
fresh	none	340
+3 months ~22°C	starting	400
+3 months 12°C	strong	900
40 h at 4°C+2 d RT	none	900
+3 months ~22°C	starting	450

Figure 7 shows the decrease of reducing sugars by reconditioning Bintje potatoes stored at 4°C from October to the beginning of March. Upon cooling, the sum of the glucose and fructose contents increased from 0.62 to 6.9 g/kg fresh weight. On day three of reconditioning at ambient temperature, this level was unchanged. After 10 and 18 days, it decreased by 46 % and 53 %, respectively: it was halved, but still remained well five times above the level before cooling. Reconditioning for 33 days reduced the sugars by 79 % (double of initial content), but such long reconditioning at room temperature easily results in unacceptable sprouting or other deterioration.

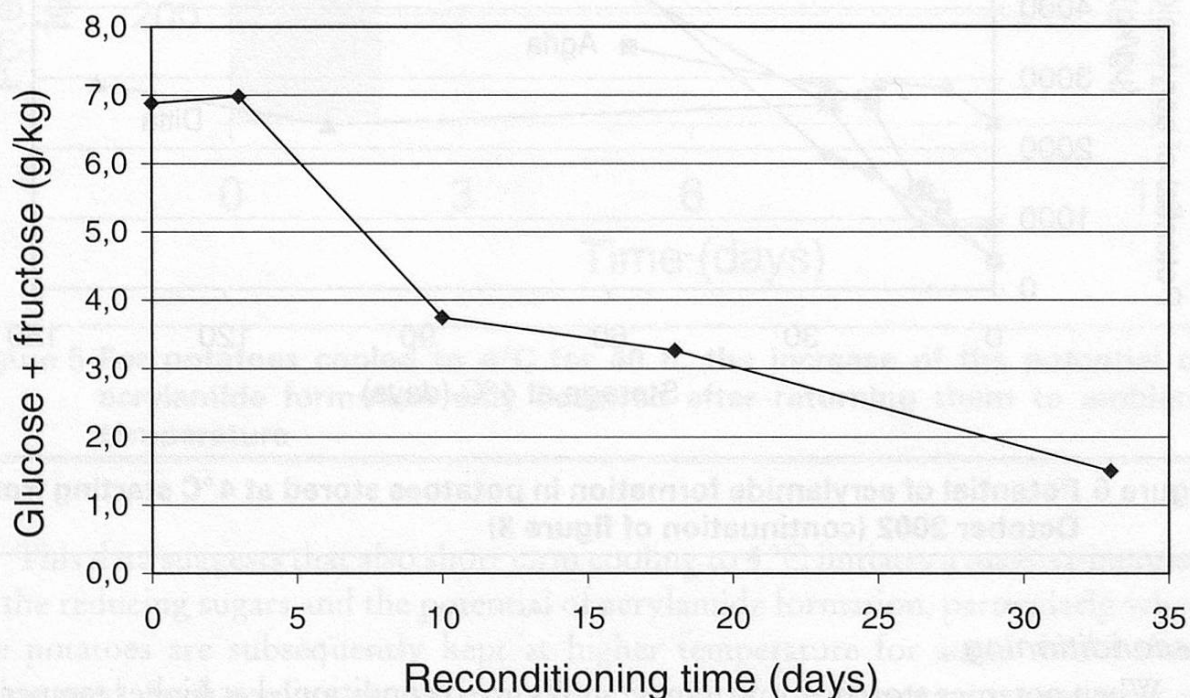


Figure 7 Reconditioning of Bintje potatoes at ambient temperature after 5 months of cooling to 4°C

Potatoes from the market

From December 2002 to March 2003, potatoes were purchased in the most important retail shops in Zurich in order to obtain data on the actual state of the potatoes offered to the consumers. Table 2 shows results on the potentials of acrylamide formation and reducing sugars. The data in the column of the fresh (non-cooled) potatoes are estimates from various results, including those from ref. (9), but were not satisfactory for several cultivars. In December, the potentials were close to or above 1000 µg/kg (average of 1480 µg/kg). At least for Bintje, Charlotte and Urgenta this means a strong increase compared to the fresh potatoes, suggesting that these samples were already from low temperature storage. In January, the potentials further increased: all except one exceeded 2000 µg/kg, and the average reached 3690 µg/kg. This means that probably all samples came from low temperature storage. Results obtained in February and March no longer changed significant (average in March: 3860 µg/kg). Contents of reducing sugars ranged from 2.2 to 14 g/kg, which is far above those found in non-cooled samples of the same cultivars (9).

Table 2

Potentials of acrylamide formation (AA) and concentrations of glucose and fructose (G+F, referring to fresh weight) in potatoes from the Swiss market purchased in December 2002 to March 2003

Cultivar	fresh	Purchasing date					
		05.12.2002	31.01.2003	13.02.2003	G+F	17.03.2003	
	AA	AA	AA	AA	G+F	AA	G+F
	(µg/kg)	(µg/kg)	(µg/kg)	(µg/kg)	(g/kg)	(µg/kg)	(g/kg)
Agata				6590	14.1		
Agria	200		1000			2260, 2310	2.7, 6.2
Appell Bio			3270			3580	8.2
Bintje	380	1070, 1320	3750, 2140	2660	2.6	3200	4.8
Charlotte	200	2855		2240	2.2	5480, 4720	11.5, 7.9
Desiree	1160	870	5990			5320	5.9
Ditta	1600		2240, 2280	3934	11.6	5820	7.8
Nicola	1000	807, 1140	3890, 3590	2730	6.4	3690, 1900	6.7, 5.2
Nicola Bio	1000		5100, 4270				
Sirtema	1000	2150	6560	4400	7.9		
Stella		2330	3060, 3260				
Urgenta	400	800	5010			4130	

Conclusions

Cooling fresh potatoes to 4°C increases their potential of acrylamide formation many fold – up to 40 times for a potato starting at a low level – which is a consequence of liberated reducing sugars. If potato products are submitted to equal heat treatment (roasting, baking, frying), this causes a correspondingly higher acrylamide content. Since these products are the main source of acrylamide for a large part of the population, cooling of potatoes below approximately 10°C is a potential health issue.

Long term storage of marketed raw potatoes largely occurs at 4°C. During February–March 2003, the average potential of acrylamide formation of potatoes from the Swiss market was above 3700 µg/kg and the mean content of reducing sugars reached 7.1 g/kg fresh weight (3.5 % referring to dry weight). This is far above the level considered suitable for frying, baking or roasting. For the preparation of potato chips, the reducing sugars should probably be below some 0.2 g/kg, for French fries below about 0.5 g/kg (10), and for roast potato (Rösti) between 0.5 and 1.5 g/kg (in preparation). It means that consumers cannot find potatoes suitable for preparing good quality (e.g. crispy) fried, roasted or baked potato products with a low acrylamide content during a long period of the year.

Storage at 8–10°C should be sufficient for most of the year, provided preference is given to suitable cultivars. Problems remain for potatoes consumed from April to the new harvest. For this critical period, cooling to lower temperatures must be evaluated against the use of chemical sprout inhibitors, i.e. acrylamide formation against the use of chemical agents. The latter may also increase the sugar contents (and even the asparagine), but the effect was reported to be far smaller than that of cooling (19). Experts in storage of potatoes should revisit the presently applied conditions with all the logistic consequences.

Cooling of potatoes occurs or is applied also in other instances. Two days of cooling at 4°C are sufficient to initiate a substantial increase of acrylamide formation. Hence even short term exposure to cold weather (farming, transport) should be avoided, just as storage in cooling rooms of distributors and restaurants, or in refrigerators at homes (together with other fresh products).

Not all cooling in the gastronomy enhances acrylamide formation. Cooling of intermediate products, e.g., peeled and cut potatoes, for 1–2 days has no significant effect as long as these are fried, baked or roasted without warming up for an extended period of time. Cooling of cooked potatoes has no effect on acrylamide formation, because the enzymes liberating reducing sugars are inactivated. Experiments also confirmed that deep freezing of potatoes has no effect on sugars and potentials of acrylamide formation, i.e. there is no objection against using frozen prefabricates, e.g. for the preparation of French fries.

Summary

Fresh potatoes respond to cooling below about 10°C by increasing the content of reducing sugars. The potential of acrylamide formation increases approximately proportionally and results in high acrylamide concentrations in fried, roasted or baked potato products. The increase at 4°C may reach a factor of 40 for a potato starting out at a low level. Cooling for merely 40 h set the signal to increase the sugar content three times (even though the increase only occurred during the following two days). Data on the increase of acrylamide formation upon medium or long term cooling to 4°C is shown for a number of cultivars frequently used in Switzerland. Reconditioning at ambient temperature for about two weeks reduced

the content of reducing sugars by a factor of two – but still left it about five times above the level before cooling. Sprouting does not noticeably increase the potential of acrylamide formation. Potatoes from the Swiss market of January to March 2003 showed potentials of acrylamide formation and sugar contents indicating that probably all samples were from low temperature storage.

Zusammenfassung

Kartoffeln reagieren auf Kühlung unter ca. 10°C mit der Freisetzung von reduzierenden Zuckern. Das Potential zur Acrylamidbildung steigt ungefähr proportional und führt zu hohen Acrylamidgehalten in frittierten, gerösteten oder gebackenen Produkten. Der Anstieg bei 4°C kann für eine Kartoffel mit einem tiefen Ausgangswert einen Faktor 40 erreichen. Kühlung während 40 h genügte, um eine Verdreifachung der reduzierenden Zucker zu induzieren, auch wenn erst in den folgenden zwei Tagen. Daten zum Anstieg der Acrylamidbildung während kurzer und langer Lagerung bei 4°C werden für in der Schweiz häufig verwendete Kartoffelsorten gezeigt. Rekonditionierung während ca. zwei Wochen bei Raumtemperatur halbierte die Zuckergehalte wieder – belies den Gehalt jedoch immer noch um einen Faktor 5 über dem Niveau vor der Kühlung. Auskeimen führt nicht zu einem bedeutenden Anstieg der Acrylamidbildung. Kartoffelproben vom Schweizer Markt von Januar bis März 2003 zeigten Potentiale zur Acrylamidbildung und Zuckergehalte, die für Lagerung bei tiefen Temperaturen typisch sind.

Résumé

Les pommes de terre réagissent au froid en libérant des sucres réducteurs dès que la température descend en dessous de 10°C environ. Le potentiel de formation d'acrylamide augmente approximativement de manière proportionnelle et engendre une teneur élevée dans les produits à base de pomme de terre frits, rôtis ou cuits au four. A une température de 4°C, l'augmentation peut atteindre un facteur de 40 pour les pommes de terre ayant une valeur basse au départ. Une réfrigération de 40 h suffit pour induire un triplement de la teneur en sucre réducteur qui s'établit dans les deux jours suivants. Des données sur l'augmentation de la formation d'acrylamide lors de périodes de réfrigération courtes et longues ont été établies sur des variétés de pommes de terre utilisées couramment en Suisse. Un reconditionnement à température ambiante d'une durée approximative de deux semaines réduit la teneur en sucres d'un facteur de deux. Celle-ci reste cependant cinq fois supérieure du niveau d'avant la réfrigération. La germination n'augmente pas de manière significative le potentiel de la formation d'acrylamide. Tous les échantillons de pommes de terre prélevées sur le marché suisse de janvier à mars 2003 présentent un potentiel de formation d'acrylamide et une teneur en sucres réducteurs typique d'un entreposage réfrigéré à basse température.

Key words

Storage of potatoes, temperature of storage, acrylamide formation, reducing sugars

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