RESEARCH ARTICLE Asian Journal of Dairy and Food Research, Volume 43 Issue 1: 32-38 (March 2024)

Evolution of Some Biochemical Parameters during Cold Storage and Their Effect on the Formation of Acrylamide in Fried Potatoes

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10.18805/ajdfr.DRF-348

ABSTRACT

Background: The province of Ain Defla (Algeria) is known for its high potato production potential. However, many challenges need to be overcome, among them the deterioration of biochemical and organoleptic quality after harvest and during storage of potato tubers.

Methods: A study was carried out in this context over two successive years 2019 and 2020, with the aim of determining the degree of physiological evolution of tubers of four potato varieties (Kondor, Bartina, Spunta and Atlas) through biochemical indicators, such as pH, acidity, dry matter, reducing sugars and starch, during cold storage at a temperature of 6°C±2 and for 60 days and 90 days. A qualitative analysis of acrylamide formation under the effect of temperature and frying time was carried out.

Result: Kondor revealed the highest pH value before and during storage. A progressive decrease in dry matter and starch as a function of storage time was detected for all varieties studied, notably Spunta and Atlas. The Spunta variety showed significant amounts of reducing sugars during 60 days of storage. Acrylamide formation was intense at 160°C for 4 minutes of frying in both Spunta and Kondor varieties. However, Kondor showed greater resistance to enzymatic browning (acrylamide) than Spunta under the same empirical conditions.

Key words: Acrylamide, Biochemical indicators, Cold storage, Potato, Variety.

INTRODUCTION

Potato tubers are rich in starch, high-quality protein and high-level vitamins. Higher levels of free amino acids and reducing sugars in potato tubers are not good for processing (Emragi *et al.,* 2022). High post-harvest perishability is one of the biggest problems in the potato tuber production chain (Bueno Da *et al.,* 2022). The use of cold storage is essential in hot-climate countries, to ensure the long-term preservation of tubers for consumption and seed potatoes (Emragi *et al.,* 2022). Fears related to the formation and accumulation of acrylamide in foods, particularly fried and cooked potatoes, have led to greater attention being paid to its toxicity. To date, most toxicity research has focused on cancerogenesis, genotoxicity, neurotoxicity and reprotoxicity (Carrara, 2018).

In Algeria, the potato sector is considered strategic. Over the years, it has acquired significant economic and social weight. Production multiplied between 2000 and 2020, reaching five million tonnes in 2020 on an area of 150 thousand hectares (Bessaoud and Lefki, 2018). The province of Ain Defla is known for its high potential for potato production, which reached 2.45 million tons over an area of 7.591 Ha. Nevertheless, many constraints remain to be overcome such as the deterioration of the biochemical and organoleptic quality of potato tubers after harvest and during storage.

This is the background to our study, whose aim is to monitor the evolution of a number of biochemical parameters, such as reducing sugars, starch, dry matter, acidity and pH in tubers of four potato varieties (red-skinned and white-skinned) intended for consumption and stored at a temperature of 6°C± 2. A qualitative analysis of the formation of acrylamide under the effect of the content of reducing sugars, the temperature and the cooking time was carried out.

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How to cite this article: Abed, A., Kaci, Z., Dahmane, T., Tirchi, N. and Cherif, D.B. (2024). Evolution of Some Biochemical Parameters during Cold Storage and Their Effect on the Formation of Acrylamide in Fried Potatoes. Asian Journal of Dairy and Food Research. 43(1): 32-38. doi: 10.18805/ajdfr.DRF-348.

Submitted: 02-08-2023 **Accepted:** 22-12-2023 **Online:** 02-01-2024

MATERIALS AND METHODS

Representative samples of potato tubers are taken from cold rooms chosen beforehand in different municipalities of the province of Ain Defla during two years, 2019 and 2020. Four varieties intended for consumption were taken into account; two varieties with white skin (Spunta and Atlas) and two varieties with red skin (Kondor and Bartina), stored for different durations (60 days and 90 days) in containers at 6°C±2 and 90% humidity. The samples are taken from containers located at the periphery and in the middle.

Preparation of potato extract for analysis

For each variety, during different storage times, five tubers are taken for the preparation of the potato extract. 100 g of tuber flesh is weighed and then ground in 150 ml of distilled water for the less sweet red-skinned varieties and in 500 ml of distilled water for the sweeter white-skinned varieties. The mixture is then filtered to obtain a potato extract.

Acidity and pH determination

The pH of the potato extract is measured using a pH meter. Total acidity, represented by total citric acid in 10 ml potato extract, is titrated with 0.1 N sodium hydroxide.

Determination of dry matter

The dry matter content of the tubers is measured after total elimination of the water content at a temperature of 70°C for 72 hours. Weights are taken before and after drying (Thompson, 1984).

Spectrophotometric determination of reducing sugars

Glucose and fructose are determined using the phenol-sulfuric acid method Dubois *et al.* (1956). Results are read at a wavelength of 480 nm for glucose and 630 nm for fructose.

Spectrophotometric determination of starch

Determination of starch content requires conversion with hydrochloric acid. Absorbance is 580 nm for polymers amylose and amylopectin and 720 nm for amylose (Browne and Zerban, 1941).

Qualitative determination of acrylamide

To detect the presence of acrylamide in potato tubers, a qualitative analysis is carried out using the non-enzymatic browning (Maillard reaction) that occurs after potato frying. Based on the results of the reducing sugar analysis, two varieties are considered: Spunta (white skin) and Kondor (red skin) as a control. Potato tuber slices with a surface area of 4cm^2 are used for frying. First, the potato slices are fried at different temperatures: 100°C, 120°C, 140°C and 160°C, with a frying time of 4 min. In a second step, the frying temperature is fixed at 120°C, with the time varying between 2 min, 3 min and 4 min. The surface areas of the brown spots on the potato slices are calculated.

All analyzes were carried out in the "Water, Rock and Plants" research laboratory at the University of Khemis-Miliana.

Analysis methods

An analysis of variance of all the biochemical parameters studied is carried out using SPSS.20.

RESULTS AND DISCUSSION

pH

In the light of the results of the statistical analysis of the Newman-Keuils test shown in Fig 1, the evolution of pH during cold storage is under the effect of the variety factor in the first order and the interaction between the variety factor and the duration of storage to a lesser extent. There were highly significant differences between the pH values obtained at variety (p=0,004) level and their interaction with storage duration (p=0,005), particularly that of 90 days. Tubers show a tendency towards a slight increase in pH over 60 and 90 days (Fig 1). However, a remarkable increase in pH during storage is observed in the white-skinned varieties Spunta and Atlas. pH is an important physiological characteristic of a tissue, which is related to the level of acidity/alkalinity due to the release or absorption of hydrogen ions (Sunmola and Bukoye, 2011). According to the same authors the increase in pH during cold storage can be attributed to a decrease in organic acids due to respiration. Juice pH is negatively correlated with the reduction in tuber sugar levels. Indeed, this result can only emphasize the influence of temperatures above 4°C on the pH increase. Khorramifar *et al.* (2022), concluded that pH variations increased slightly during the storage period, compared to

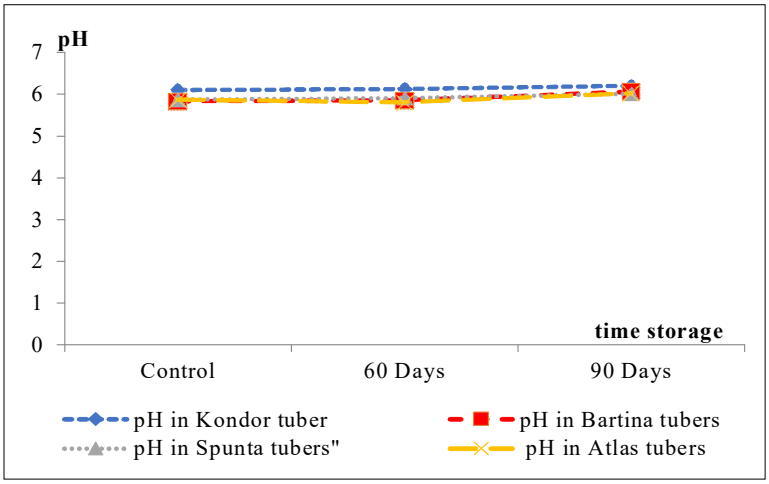


Fig 1: Evolution of the pH (within p<0.05) during times storage at 6°C±2.

that at harvest which was found to be slightly lower than the storage period.

Acidity

Statistical analysis of changes in acidity during storage reveals highly significant differences in terms of variety (p=0,006), storage time and interactions between the two (p=0,003). In fact, the values generated in Fig 2 show a certain gain in acidity for the Kondor variety compared with the other varieties studied. A slight increase in acidity was revealed during both storage periods, with an greatest for the 90 days period in the white skinned Spunta and Atlas followed by the Bartina variety. According to Poberężny (2006), organic acids in potato tubers total around 0.4-1.0% of their fresh weight citric acid concentrations depend mainly on genetics, which is supported in this study. Sulaiman (2005) and Poberężny (2006) found higher citric acid content after 6 months' storage than after harvest.

Dry matter

Analysis of variance reveal highly significant differences in the evolution of dry matter under the effect of variety (p=0,004), but especially under the effect of storage duration (p=0,001). In fact, average dry matter values are higher in control tubers than in stored tubers for all the varieties studied (Fig 3). A regression in the average value of this parameter is observed during both storage periods, particularly the 90 day period. It seems that the red-skinned varieties Kondor and Bartina are more resistant to storage conditions than the white-skinned varieties (Spunta and Atlas). These results corroborate those reported by Abdelgaleel *et al.* (2006) who deduced that tuber dry matter content was significantly affected by storage conditions and storage period. De Freitas (2012) concluded that dry matter content decreased during storage for all potato clones and that lower storage temperatures (4 and 8°C) tended to be more effective in maintaining it. While Nantongo *et al.* (2023), reported that dry matter content, all origins, increased from 0 to 45 days' storage and decreased at 60 days' storage. Transpiration is responsible for around 90% of the total loss. In comparison, weight loss due to respiration accounts for less than 10% of total loss (Emragi *et al.,* 2022).

Starch (Amylose and amylopectin)

Statistical analysis of the mean values of the two starch components, amylose and amylopectin, reveals highly

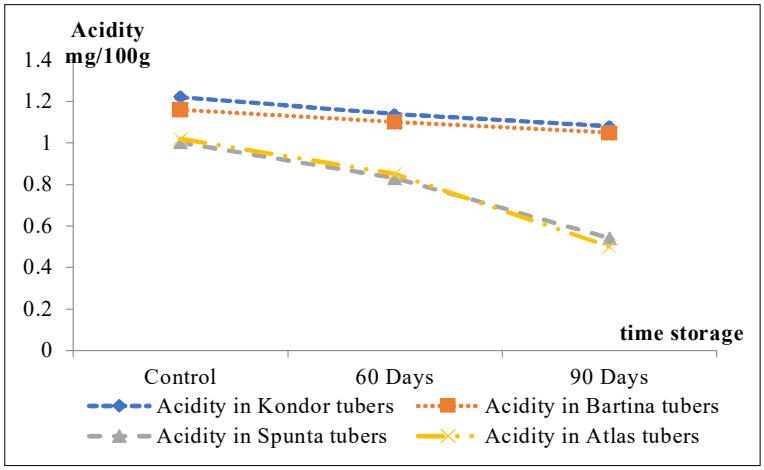


Fig 2: Evolution of the acidity (mg/100 g) (within p<0.05) during times storage at 6°C±2.

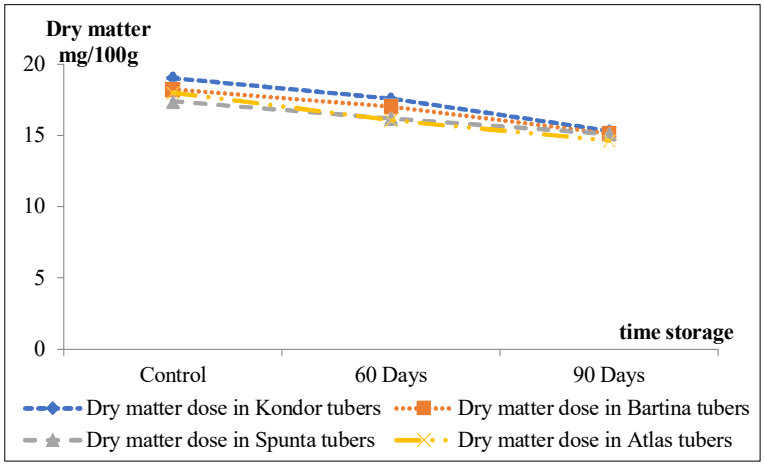


Fig 3: Evolution of the dry matter (mg/100 g) (within p<0.05) during times storage at 6°C±2.

significant differences between the varieties studied (p=0,003) and between them and the duration of storage at 6°C±2 (p=0,002). A sharp decrease in amylose and amylopectin is recorded for all cultivars during 60 and 90 days of storage and more markedly for the white-skinned varieties Spunta (4.5, 13.8 g/100 g) and Atlas (5.05, 18.5 g/100 g) (Fig 4).

El-Sayed *et al.* (2007) have reported that the Spunta variety performs less well in cold storage than the Diamant variety, as its tubers contain higher levels of starch and total phenols. It could therefore have an active mechanism by which it slows down or inhibits the activity of degradable and respiratory enzymes. According to Lahmar (2018), the effect of the storage period has been well indicated on the evolution of the starch content with its two components (amylose and amylopectin), the latter decreasing as the storage period increases. In the same context, Siddiqui *et al.* (2022) deduced that the starch content of potatoes decreases during storage due to the conversion of starch into sugar and its use in respiration, with a substantial reduction in the amylose content of the starch with increasing tuber storage time.

Although levels of these two components were lower before storage in the Kondor and Bartina varieties, this did not affect levels at the end of storage, compared with Spunta and Atlas, which had higher levels before storage (Fig 4). The starch content of tubers is a function of genetic and environmental factors (De Freitas, 2012). Abbasi *et al.* (2016), deduced from their results that the significant decrease in starch content at the lowest storage temperatures (5°C to 6°C) could be associated with higher invertase activity. According to the same author, the significant decrease in starch content is largely a function of storage temperature. These results were similar to those obtained in this study. According to Cruz *et al.* (2021), cold is also an abiotic stress for potato tubers, which can induce enhanced starch-sugar metabolism and cause sugar accumulation.

Reducing sugars (Glucose and fructose)

The results of the statistical analysis shown in Fig 5 reveal a sharp increase in glucose and fructose content in all the cultivars studied. Highly significant differences are also recorded between varieties (p=0.002) and between varieties and storage time (p=0.001). A marked increase in glucose and fructose content is observed in the white-skinned varieties Spunta and Atlas, respectively, during the various storage periods, particularly the 90 day period, compared with Kondor and Bartina (Fig 5). It seems that the decrease in starch over the two storage periods is inversely proportional to the glucose and fructose content in all cultivars. In addition, varieties with low dry matter content are more likely to have higher reducing sugar contents, as is the case with Spunta and Atlas respectively. Matsuura-Endo *et al.* (2006) reported that tuber sugar content increased significantly at temperatures below 8°C for all cultivars, even after less than 2 weeks' storage. Our results show that the influence of storage time on glucose and fructose content is more pronounced than that of storage temperature. According to Salomão *et al.* (2022), tubers stored at ambient temperature had tens of times lower levels of reducing sugars than those stored at 5°C or 6°C. Cruz *et al.* (2021) detected higher levels of total reducing soluble sugars in tubers, of two cultivars, stored at 6°C with a peak at 30 days of storage and a reduction in the other periods. The increase in reducing sugars in tubers stored at 6°C after 30 days is due to increased invertase. The high levels of reducing sugars in both Spunta and Atlas reflect a trait of great importance when it comes to the destination of these two cultivars for industrial processing.

Evolution of non-enzymatic browning in stored potatoes

Evolution of non-enzymatic browning as a function of frying temperature

Highly significant differences are recorded between the two varieties studied, Spunta and Kondor and the interaction

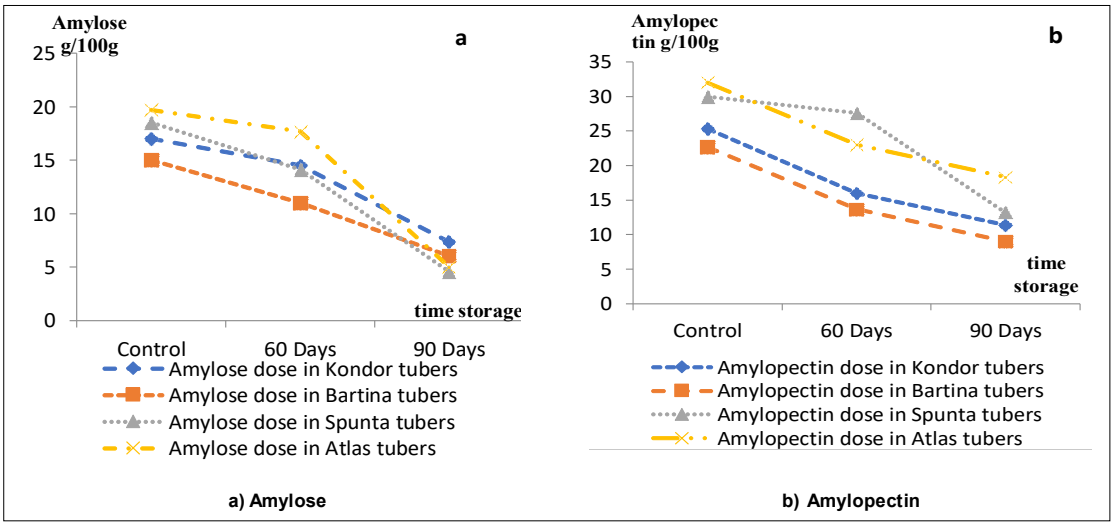


Fig 4: Evolution of the amylose and amylopectin contents (within p<0.05) during times storage at 6°C±2.

between them, storage time and frying temperature, which vary between 100°C, 120°C, 140°C and 160°C, with frying time set at 4 min. The average values for the appearance of brown surfaces after frying fluctuate considerably, depending on the variety stored for 60 days and the frying temperature. The increase in frying temperature leads to an increase in the brown surface area of the slices potato, 4 cm^2 at 160°C; this means that the entire surface of the slices is brown. The effect of the reducing sugar content has a strong impact on the appearance of brown spots on the slices fried. In this respect, the Spunta variety prove to be very sensitive to frying temperature due to its high glucose and fructose content, compared with the Kondor variety, above 100°C (Table 1).

Evolution of non-enzymatic browning as a function of frying time

By setting the frying temperature at 120°C and varying frying times from 2 min, 3 min to 4 mn, highly significant differences appear between stored varieties and their interactions with frying times. The mean values for brown spots on cooked potato slices are highly indicative of the susceptibility of the Spunta variety, stored at 60 days, to heat for 4 minutes, followed by the Kondor variety. Apart from the 4mn frying time, which is of particular importance in the appearance of brown spots, the 2 mn and 3 mn frying times have no effect (Table 2).

According to Stojanovska and Tomovska (2015), storage temperature strongly influences reducing sugars, identified as the most important precursors for acrylamide formation. The main limiting factors responsible for acrylamide formation in potato products are respectively reducing sugars (glucose and fructose) and free asparagine. Acrylamide formation occurs mainly under conditions of high temperature (generally above 120°C) and low humidity. For Halford *et al.* (2022), the ratio of free asparagine to reducing sugars in potatoes means that the concentrations of the latter generally determine the amount of acrylamide formed in potato products. However, the concentration of free asparagine can contribute to acrylamide variance, particularly in varieties with relatively high concentrations of reducing sugars.

Matsuura-Endo *et al.* (2006) concluded that reducing sugar content increases markedly in all cultivars at

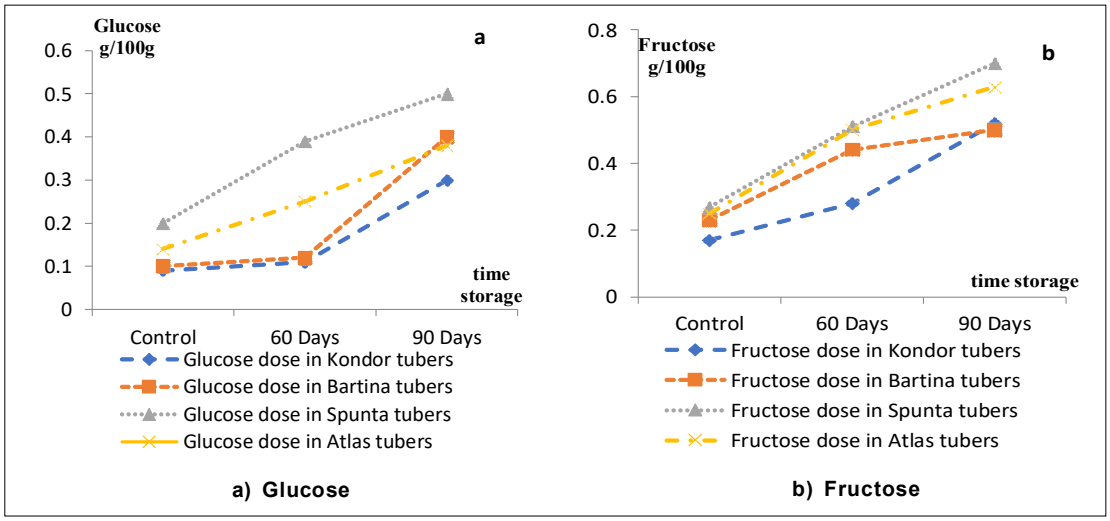


Fig 5: Evolution of the glucose and fructose contents (within $p<0.05$) during times storage at 6°C±2.

Table 1: Evolution of brown spots in cm^2 on slices of stored tubers according to frying temperatures.

Variety	Control				60 days			
	100°C	120°C	140°C	160°C	100°C	120°C	140°C	160°C
Kondor	0	0	0	0	0	0.42	0.47	1
Spunta	0	0	0	0	0	3.41	3.6	4

Table 2: Evolution of brown spots in cm^2 on slices of stored tubers according to frying times.

Variety	Control			60 days		
	2 mn	3 mn	4 mn	2 mn	3 mn	4 mn
Kondor	0	0	0	0	0	0.43
Spunta	0	0	0	0	0	3.41

temperatures below 8°C, with similar increases in acrylamide levels and dark brown slices color. Free amino acids changed little at the storage temperatures tested. Reducing sugar content correlated well with acrylamide level when the fructose/asparagine molar ratio in tubers was below 2. When the fructose/asparagine ratio was above 2 for low temperature storage, asparagine content, rather than reducing sugar content, was proved to be the limiting factor in acrylamide formation.

CONCLUSION

The study of the behavior of four potato varieties (Kondor, Bartina, Spunta and Atlas) under the effect of cold storage at 6°C±2 put an accent on the destination of each variety either for consumption or industrial processing through the evolution of some biochemical parameters. It seems that the white-skinned varieties Spunta and Atlas are more sensitive to cold storage than the red-skinned varieties Kondor and Bartina. In fact, trends towards decreasing or increasing biochemical parameters such as pH, acidity, dry matter, reducing sugars and the appearance of non-enzymatic browning are observed to a greater or lesser extent in Spunta and Atlas tubers than in Kondor and Bartina. In this case, the evolution of pH and acidity, with a slight increase in both Spunta and Atlas varieties, is under the effect of variety rather than storage conditions or duration. It should also be noted that the sharp increase in reducing sugars during the 60 day and 90 day storage periods has a strong impact on the appearance of brown spots on the surfaces of white-skinned potato slices, due to non-enzymatic browning. The decrease in dry matter content is closely linked to the degradation of starch with its two components amylose and amylopectin.

Conflict of interest

We wish to confirm that there are no known conflicts of interest associated with this publication and there has been no significant financial support for this work that could have influenced its outcome.

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