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Reducing the acrylamide concentration in homemade bread processed with L-asparaginase

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ABSTRACT

Acrylamide is the main contaminant for starchy foods cooked at temperatures above 120 °C. It represents a potential carcinogen, and consequently, its reduction is important for safeguarding global health. The L-asparaginase enzyme catalyses the hydrolytic cleavage of L-asparagine, a precursor of acrylamide, into L-aspartic acid and ammonia, thus reducing the formation of acrylamide. The objective of this work was to test the enzyme L-asparaginase at two concentrations (150 and 300 U/kg flour) under normal baking conditions to reduce acrylamide. The results showed that the use of 300 U/g of the enzyme led to a reduction of 78% in acrylamide, meeting the reference level reported by **Reg. (EU) 2017/2158** (50 μg/kg) without influencing parameters such as colour that impact the final characteristics of the product.

1. Introduction

Bread and bakery products represent important foods in the human diet worldwide (Melini, Vescovo, Melini, & Raffo, 2024). The Maillard reaction (MR) is a reaction that occurs during the thermal treatment of foods and influences their flavour, aroma, and colour (Pedreschi, Mariotti, Granby, & Risum, 2011). Unfortunately, starch-rich processed foods such as bread also produce acrylamide, a harmful product (Teng, Hu, Tao & Wang, 2018). Acrylamide (AA; $CH_2 = CHCONH_2$) has been classified as a foodborne toxicant since 2002 for its genotoxic, carcinogenic, and neurotoxic ability (Abedi, Hashemi, & Ghiasi, 2023; Augustine & Bent, 2022), and it has become an increasingly important topic in food safety in recent years. Therefore, in European countries, the European Food Safety Authority (EFSA) has estimated a dosage range within which acrylamide is likely to cause a slight but measurable incidence of cancer or other adverse effects (related to neurological processes, pre- and postnatal development and the male reproductive system). The lower limit of this range is called the "*lower limit of the reference dose confidence interval*" ($BMDL_{10}$). For tumours, experts have established a $BMDL_{10}$ of 170 μg/kg body weight (b.w.) per day, whereas for other effects, the value is 430 μg/kg body weight (b.w.) per day. According to the EFSA, this value should be used as a reference point for calculating the margin of exposure (MOE) (EFSA, 2015a). The calculated MOEs of acrylamide are 425 and 1075, for cancer risk and other undesirable effects in adults, respectively, and 50 and 126 in children (EFSA, 2015b). The formation of acrylamide occurs mainly through the reaction between free L-asparagine and reducing sugars when the moisture content of the product is less than 30% and the cooking temperature is above 120 °C (Mollakhalili-Meybodi, Khorshidian, Nematollahi, & Arab, 2021). Thus, acrylamide levels in heat–processed foods depend on various factors, including the recipe (initial precursor concentrations, ingredients, additives) and processing conditions (processing time and temperatures, pH, humidity, physical parameters, heat transfer) (Rifai & Saleh, 2020; Maan et al., 2022). Various strategies exist for reducing acrylamide levels in these foods due to its impractical removal from prepared food products (Sarion, Codină, & Dabija, 2021). For example, Przygodzka et al. (2015) reported that prolonged cooking at lower temperatures can reduce acrylamide levels. The addition of complexing agents, such as polyvalent cations, may inhibit acrylamide formation by forming bonds with L-asparagine (Pedreschi, Granby, & Risum, 2010). Moreover, the addition of amino acids such as glycine can reduce acrylamide formation by competing with L-asparagine in the Maillard reaction (Zhu et al., 2016). In this context, enzymatic treatments stand out as simple and effective methods for reducing acrylamide without affecting the nutritional and sensory properties of the final product (Jia et al., 2021). Consequently, research

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has focused on strategies to mitigate and limit acrylamide formation in foods (Hendriksen, Kornbrust, Østergaard, & Stringer, 2009) without compromising their sensory and nutritional characteristics. Gazi, Göncüoğlu Taş, Görgülü, and Gökmen (2023) reported that for effective enzymatic treatment, controlling key parameters such as the enzyme dose, reaction time, temperature, and water activity is crucial. Additionally, the enzyme dosage must not alter the rheological and textural properties of the final products (Rosell, Rojas & De Barber, 2001). Bakery goods can contribute to high levels of acrylamide exposure due to their high consumption. **Reg. (EU) 2017/2158** reported 50 μg/kg as a reference level of acrylamide in soft bread, while the content of AA in baked goods and bread on the market can vary from 31 to 454 μg/kg, as reported by Esposito, Velotto, Rea, Stasi, and Cirillo (2020). To reduce the presence of AA in baked goods, the food industry must change the processes and parameters of the product without compromising taste, texture and appearance. The Food Industry Confederation "FoodDrink Europe", in close cooperation with the National Authorities and the European Commission (EU), has developed the *Acrylamide Toolbox* that helps food business operators identify the best ways to reduce acrylamide in their products as acrylamide has been considered by authorities as a major food emergency (FDE, 2019). This toolbox is a useful tool to properly implement the **Reg. (EU) 2017/2158** and reducing the levels of acrylamide in food to "as low as is reasonably achievable" (ALARA principle). Several methods for acrylamide mitigation have been studied in the literature, including heat treatments such as blanching (Zhang, Kahl, Bizimungu, & Lu, 2018), fermentation by lactic acid bacteria or yeasts (Akıllıoglu & Gökmen, 2014; Nachi et al., 2018), reduction of temperature−time treatment (Bachir, Haddarah, Sepulcre, & Pujola, 2022), treatment with L-asparaginase (Covino et al., 2023; Dias, Junior, Hantao, Augusto, & Sato, 2017; Sharma & Mishra, 2017), and replacement of ingredients such as sugars (Abdel–Shafi, Wei–ning, Zhen–ni, & Chung, 2011). To mitigate the formation of acrylamide in baked goods without compromising the organoleptic and sensory quality of the final product, the use of the enzyme L-asparaginase is a powerful strategy for the food industry (Xu, Oruna-Concha, & Elmore, 2016). This enzyme can hydrolyse the amino acid L-asparagine present in the farinaceous in a wide range – from approximately 7 mg–70 mg/100 g (Alam, Ahmad, Pranaw, Mishra, & Khare, 2018; Stockmann et al., 2018). L-asparaginase has been recognized as a GRAS by the Food and Drug Administration (FDA). To the best of our knowledge, studies in the literature on the use of L-asparaginase in the production of homemade bread but not sweet bread (Kumar & Bachhawat, 2012; Meghavarnam & Janakiraman, 2018), special bread (Capuano et al., 2009; Ghorbani, Tehrani, Khodaparast, & Farhoosh, 2019), or similar products (Anese, Quarta, & Frias, 2011; Gazi et al., 2023) are limited. Therefore, the aim of this work was to mitigate acrylamide through the use of L-asparaginase in homemade bread. In addition, this study evaluated the possibility of using the enzyme under its suboptimal conditions that occur during baking. The acrylamide content, reducing sugars, L-asparagine content, water activity, moisture content and colorimetric indices were determined to evaluate the effects of two different concentrations of the L-asparaginase enzyme (150 and 300 U/kg flour) on acrylamide mitigation and macroscopic parameters.

2. Materials and methods

2.1. Chemicals and raw materials

All solvents and reagents used for the experiments were purchased from Sigma–Aldrich Co. (Milano, Italy). The enzyme L-asparaginase Acrylaway 3500 BG® (3500 ASNU/g) from Novozymes A/S (Bagsvaerd, Denmark) produced by *Aspergillus oryzae* (optimal pH 7 and optimal temperature 37 °C) was used. The ingredients used to prepare the breads were purchased from a local supermarket: wheat flour "00" (Molino Naldoni, Italy) and fresh yeast *Saccharomyces cerevisiae* (Lievital, Italy), sodium chloride (Italkali, Italy) and water.

2.2. Bread preparation

The recipe was formulated with 58.4% flour, 38.9% water, 1.5% yeast, and 1.2% salt. In a planetary mixer (Howell deluxe mixer - HIMP810), fresh brewer's yeast (approximately 3% of the flour weight) was dissolved in water maintained at room temperature (25 °C). The enzyme L-asparaginase was dissolved in water at various concentrations, 0 (control), 150 U, and 300 U, to ensure better homogeneity in the dough. The enzyme was added to the flour to replace of the same amount of flour to maintain the overall quantity of solids in the recipe. Flour was added gradually, and finally, salt was added. The dough was kneaded at medium speed for 15 min. The obtained doughs, both treated and untreated, were placed in a fermentation chamber (Panasonic MIR-154-PE Cooled Incubator, Osaka, Japan) at 37 °C for 3 h. Specifically, the process involved a first step of leavening at 37 °C for 2 h on the whole dough, followed by a second step of leavening at 37 °C for 1 h after the dough had formed. The fermentation conditions were chosen to respect the optimal temperature for both the enzyme and the yeast. After the fermentation phase, the samples were baked in an oven (BAKE OFF ITALIANA, Ferrara, Italy) at 260 °C for 40 min, with a ventilation rate of 2 out of 5. The bread samples obtained, weighing approximately 500 g each, were round in shape and had a diameter of 20–22 cm.

The samples (dough and baked bread) were frozen at −20 °C, freeze-dried for 48 h (−50 °C, <0.05 mbar), and kept in a dark and dry place.

The samples were analysed before baking (dough pre- and post-fermentation) and after baking (bread). Table 1 lists the codes of the samples used in this study.

2.3. Physicochemical analysis

After calibration, the pH values of the dough were measured with a basic pH meter 20 (Crison Instruments, Barcelona, Spain). Briefly, 15 g of each sample was dissolved in 100 mL of deionized water as reported by Park, Jung, and Jeon (2006). The moisture content (mc%) of the dough and bread was evaluated via the gravimetric method following the official method (*Gazzetta Ufficiale*) and **Equation (1)**. Briefly, 3 g of each sample were oven-dried at 105 °C for 12 h and accurately weighed until a constant weight was reached. The moisture content was expressed as a weight/weight percentage of water (% *w/w*). The a_w of the dough before and after fermentation was measured via an Aqua Lab instrument (METER Group, Inc., Pullman, WA, USA).

$$moisture\ content = \left(\frac{Mi - Mf}{Mi}\right) * 100 \qquad \text{Equation 1}$$

Calculation for mc% where:

Mi is the initial weight of the sample
Mf is the final weight of the sample.

2.4. Determination of reducing sugar content in dough

The reducing sugars in the dough were extracted following the method reported by Liyanage, Yevtushenko, Konschuh, Bizimungu, and

Table 1
Codes of samples used for the study.

Sample	Asparaginase treatment (U/kg flour)	Code
Dough Pre-fermentation	0	C-Pre
Dough Pre-Fermentation	150	150-Pre
Dough Pre-Fermentation	300	300-Pre
Dough Post-Fermentation	0	C-Post
Dough Post-Fermentation	150	150-Post
Dough Post-Fermentation	300	300-Post
Bread	0	C-Bread
Bread	150	150-Bread
Bread	300	300-Bread

Lu (2021), with some modifications. Briefly, approximately 0.5 g of lyophilized sample was weighed into a 50 mL tube and mixed with 10 mL of ultrapure water, and 1 mL of Carrez I and II was added. The sample was subsequently sonicated for 1 h, centrifuged at 6500 rpm for 10 min and filtered with a 0.22-μm PES filter (Phenomenex, Torrance, CA, USA). The obtained supernatants were collected and used for spectrophotometric analysis. The monosaccharide contents (D-glucose and D-fructose) were determined spectrophotometrically using a Megazyme Kit (D-fructose/D-glucose Kit, Megazyme®, Chicago, IL, USA) according to the manufacturer's protocol. The determinations were made at 340 nm via a UV–visible spectrophotometer (Shimadzu UV-1601PC, Milano, IT).

2.5. L-asparagine content in dough

To determine the L-asparagine content in the dough, approximately 2 g of sample was suspended in 100 mL of water, sonicated for 1 h, centrifuged at 6500 rpm for 10 min, and then filtered with a 0.22-μm PES filter (Phenomenex, Torrance, CA, USA). The L-asparagine content was determined using an L-asparagine/L-glutamine (Rapid) Assay Kit (Megazyme®), and the assays were conducted according to the manufacturer's instructions. The determinations were made at 340 nm via a UV–visible spectrophotometer (Shimadzu UV-1601PC, Milano, IT).

2.6. Acrylamide determination in bread

Acrylamide (AA) extraction and determination from bread was carried out as reported by Mohan Kumar, Shimray, Indrani, & Manonmani (2014), with modifications. Briefly, 1 g of freeze-dried ground bread was homogenized with 10 mL of water; then, 0.5 mL of Carrez A and Carrez B were added for purification. The sample was subsequently stirred in a thermic bath (Falc Instruments, Bergamo, Italy) at 70 °C for 30 min. The homogenates were subsequently centrifuged at 6.500 rpm for 15 min to allow further purification and filtered first on Whatman filter paper No. 2 and then through 0.22 μm PES filters. The AA content of the bread was determined via UHPLC (Jasco LC-4000, Tokyo, Japan) equipped with a MD-4010 PDA detector, an Oven CO-4061 column, a UHPLC Semimicro Pump PU-4285, and a C18 reversed-phase column (Nucleodur C18 Gravity 150 × 3 mm, 3 μm). The pretreated sample (10 μL) was injected into the UHPLC system. Analysis was carried out isocratically using acidified water with formic acid (0.1%) as the mobile phase, with a flow rate of 0.25 mL min^{-1}. The detector wavelength was set at 210 nm. The identification and quantification of AA was performed by comparing the retention time (4.6 min) and the area between the peak of the external standard (0.1–50 ppm, $R^2 = 0.99$) and the peaks of the samples. The results were expressed in μg kg^{-1} bread.

2.7. Colorimetric analysis

The colorimetric indices (L^*, a^*, and b^*) of the samples were measured with a portable digital colorimeter (WR10QC, Beley, China), and the CIELab coordinates (L^*, a^*, b^*) were recorded. The colour coordinate L^* gives the lightness ranging from black at 0 to white at 100. The chromaticity coordinates a^* are red (+) and green (−), whereas the chromaticity coordinates b^* are yellow (+) and blue (−). L, a, and b parameters were used to calculate the browning index (BI) through the following formula reported by Verma, Singh, Chauhan, and Yadav (2023).

$$BI = \frac{[\,100 * (X - 0.31)\,]}{(0.17)}$$

$$X = \frac{a^* + 1.75L^*}{5.645L^* + a^* - 3.012b^*}$$

2.8. Statistical analysis

All analyses and determinations were performed in triplicate, and the reported results are the average values obtained ± standard deviation. The data were subjected to one-way analysis of variance (ANOVA) and Tukey's multiple range test ($p \leq 0.05$) using XLSTAT software (Addinsoft, New York, USA).

3. Results and discussion

3.1. pH, water activity, and moisture content

The values of pH, water activity (a_w), and moisture content (mc%) are reported in Table 2. The pH value ranged from 5.29 in dough 300-post to 5.38 in dough C-Pre, 150-Pre. During fermentation, the pH decreases due to the production of organic acids (Su et al., 2019). During fermentation, the moisture content tends to decrease, probably because of the evaporation of water during this phase; however, L-asparaginase, which can work with at least 30% of the water in the matrix (Hendriksen et al., 2009), remains in favourable conditions, whereas a_w is high because of the high degree of hydration of the dough (approximately 67%). Moreover, no significant difference was found for a_w and moisture content. These factors are important factors to consider during dough production because they affect the working conditions of the enzyme L-asparaginase. The pH and a_w values of the control and enzyme-treated bread samples were not significantly different (Table 2).

The moisture content of bread samples ranged from 32.59% to 34.49%, which was in accordance with the limits reported by **Law 580/July 4, 1967.** Compared with that of dough, the moisture content (mc%) of bread is approximately 30% lower because of water evaporation during baking.

3.2. L-asparagine and reducing sugar contents

The free L-asparagine content found in the flour was 216.90 mg kg^{-1}, which was in accordance with the results of Mesias, Delgado-Andrade, and Morales (2022) and Ciesarová, Kukurová, Bednáriková, and Morales (2009). Fig. 1 shows the L-asparagine content and reducing sugar content in the dough before and after fermentation. The control dough (not subjected to enzymatic treatment) showed a reduction in the L-asparagine content of approximately 30%, from 1.39 to 0.97 g kg^{-1} during fermentation. These results are in line with those of Claus, Mongili, Weisz, Schieber, and Carle (2008) and Fredriksson, Tallving, Rosén, and Åman (2004), who studied the effects of fermentation time on the L-asparagine and acrylamide contents of bread. They reported a decrease in the L-asparagine concentration in doughs as the fermentation time increased, suggesting the possible consumption of

Table 2
Physico-chemical parameters value in dough and bread.

Dough			
Sample	pH	a_w	mc (%)
C-Pre	5.38 ± 0.01[a]	0.97 ± 0.01[a]	47.11 ± 0.21[a]
150-Pre	5.38 ± 0.02[a]	0.98 ± 0.01[a]	47.71 ± 0.34[a]
300-Pre	5.32 ± 0.01[b]	0.98 ± 0.02[a]	48.00 ± 1.12[a]
C-Post	5.33 ± 0.01[b]	0.97 ± 0.01[a]	46.71 ± 0.21[a]
150-Post	5.31 ± 0.04[bc]	0.98 ± 0.01[a]	47.20 ± 0.37[a]
300-Post	5.29 ± 0.01[c]	0.97 ± 0.02[a]	47.66 ± 0.24[a]
Bread			
Sample	pH	a_w	mc (%)
C-Bread	5.57 ± 0.02[a]	0.88 ± 0.02[a]	32.59 ± 0.81[b]
150-Bread	5.60 ± 0.03[a]	0.90 ± 0.01[a]	34.49 ± 0.88[a]
300-Bread	5.59 ± 0.01[a]	0.89 ± 0.02[a]	33.40 ± 1.76[ab]

[a-c]Different letters in the same column indicate statistically significant differences ($p < 0.05$).

[a-d]Different letters on the bars indicate statistically significant differences (p<0.05).

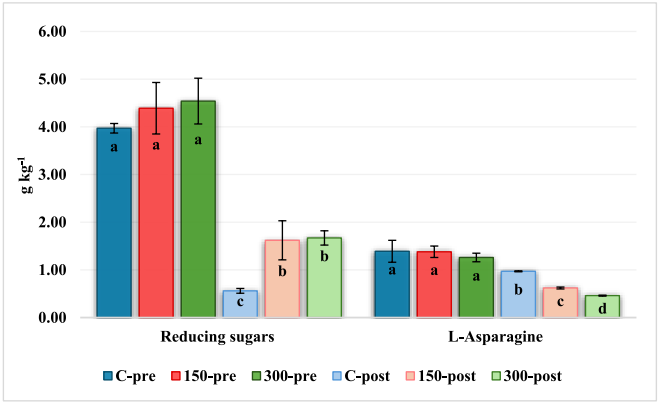

Fig. 1. Reducing sugar and L-asparagine content (g kg^{-1}) in pre- and post-fermentation dough.

[a-c]Different letters indicate statistically significant differences (p<0.05)

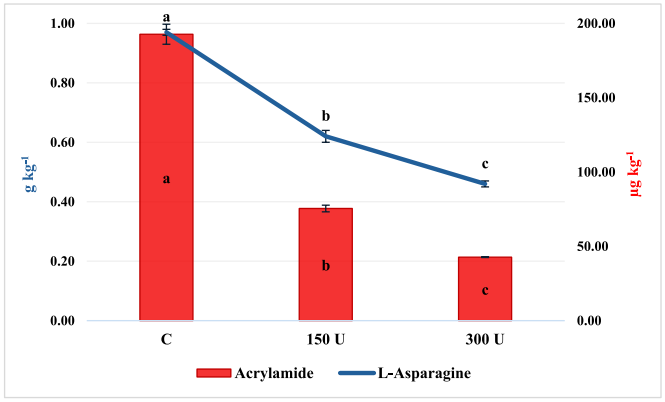

Fig. 2. Acrylamide (µg kg^{-1}) content in bread samples and L-asparagine (g kg^{-1}) post-fermentation dough samples.

L-asparagine by yeast. In the samples treated with L-asparaginase, the L-asparagine content was lower than that in the control samples (55.49% and 63.78% for the doughs treated with 150 U and 300 U of the enzyme, respectively). These findings, in accordance with those reported by Hendriksen et al. (2009), who investigated the mitigation of acrylamide content through the use of enzymatic treatments, suggest effective enzyme activity that hydrolysed L-asparagine into L-aspartic acid during fermentation. Furthermore, Gazi et al. (2023) reported a reduction in the L-asparagine content in the dough of different bakery products treated with L-asparaginase, and this reduction was greater in products with the highest water activity.

In terms of the reducing sugar content for all the samples, a clear decrease in reducing sugars was observed between pre- and post-fermentation. Indeed, the yeast *S. cerevisiae* uses sugars during alcoholic fermentation, resulting in the production of ethyl alcohol, carbon dioxide (CO_2) and heat energy (Cho & Peterson, 2010). Specifically, for the control sample (C), there was a reduction of approximately 86%, whereas for the 150-Post and 300-Post samples, the reductions in fermenting sugars were 74% and 63%, respectively. In bread, a greater reducing sugar content was detected in the control (C-bread) samples than in the enzyme-treated samples (150-Bread and 300-Bread), as shown in Table 3. This result differs from that of Mohan Kumar, N, Shimray, Indrani, and Manonmani (2014), who reported a reduction in reducing sugars in bread samples treated with L-asparaginase. The higher sugar content in the bread treated with the enzyme than in the control bread could be explained by the greater presence of L-asparagine in the non–enzyme-treated bread, which then reacts with the reducing sugars during the Maillard reaction.

3.3. Acrylamide content in bread

The acrylamide content in the baked samples, compared with the L-asparagine content in the post-fermentation dough samples, is shown in Fig. 2. In the control sample (C-Bread), an acrylamide content of approximately 193 µg kg^{-1} was detected. According to the literature, the acrylamide content in white bread typically falls within the range of 100–450 µg kg^{-1}, depending on the raw materials used, fermentation time, and baking conditions (Boyacı Gündüz & Cengiz, 2015; Wang, Yu, Xin, Wang, & Copeland, 2017). However, in the enzymatically treated samples 150-Bread and 300-Bread, the acrylamide content was reduced by approximately 61% and 78%, respectively, compared with that of the control. Specifically, bread treated with 150 U had an acrylamide content of 75 µg kg^{-1}, whereas the bread treated with 300 U had a content of 42.74 µg kg^{-1}. In addition, the decrease in acrylamide content was also related to the smaller decrease in reducing sugars in the enzyme-treated samples, which is witnessed during cooking. The 300-Bread sample complies with the reference level for acrylamide content outlined in **Reg. UE 2158/2017**, which reports a limit of 50 µg kg^{-1} for soft bread (moisture content above 10%). The use of L-asparaginase to reduce the acrylamide content in different food products has been previously reported. Dias et al. (2017) reported a reduction in acrylamide in French fries treated with L-asparaginase (50 U/mL) from *A. oryzae*. Additionally, Zuo, Zhang, Jiang & Mu (2015) reported an acrylamide content reduction in French fries when they were treated with L-asparaginase from *Thermococcus zilligii*. Furthermore, Mohan Kumar, Shimray, Indrani & Manonmani (2014) reported a reduction in the acrylamide content when the L-asparaginase level in sweet bread increased.

Table 3
Reducing sugar (g kg^{-1}) content in bread.

Sample	Reducing sugar
C-Bread	0.15 ± 0.02^{c}
150-Bread	0.27 ± 0.06^{b}
300-Bread	1.47 ± 0.12^{a}

[a-c]Different letters in the same column indicate statistically significant differences ($p < 0.05$).

3.4. Colorimetric analysis

The colour indices are reported in Table 4. The value of a*, which is a measurement of the red colour, ranged between −9.31 and −9.11, whereas the value of b*, an indicator of the yellow colour, ranged from 9.16 to 9.28.

All colorimeter indices (L*, a* and b*) were not significantly different, leading to the conclusion that the addition of the enzyme does not lead to colorimetric changes, an important aspect to consider in the development of food. Regarding the Browning Index (BI), the following values can be observed for the control bread and the bread treated with enzymes at 150 U and 300 U: 15.40, 15.24 and 14.99. There was a slight decrease in the BI with increasing enzyme units. However, the differences are not statistically significant, meaning that there is no significant impact on this index. This result is positive, as one of the objectives of

Table 4
Colorimetric parameters of bread samples.

Sample	L*	a*	b*	BI
C-Bread	21.90 ± 0.11	−9.11 ± 0.03	9.16 ± 0.07	15.40 ± 0.62
150-Bread	21.91 ± 0.84	−9.31 ± 0.21	9.28 ± 0.18	15.24 ± 1.14
300-Bread	22.51 ± 0.12	−9.12 ± 0.01	9.20 ± 0.06	14.99 ± 0.52

No statistically significant differences were found ($p > 0.05$).

using the enzyme is to preserve the sensory and quality properties of the final product (Jia et al., 2021), ensuring general consumer acceptability. Indeed, colour is one of the most valued attributes of Maillard's reaction. According to Michalak, Czarnowska-Kujawska, and Gujska (2019), there was no statistically significant correlation between colour indices and the acrylamide content of commercial bread. The use of L-asparaginase is one of the best methods to eliminate the presence of L-asparagine as well as the formation of acrylamide in baked goods and does not affect other parameters, such as browning (Abdel–Shafi et al., 2011; Anese et al., 2011).

4. Conclusion

Acrylamide is a toxic substance formed during the baking process at high temperatures (>120 °C) as a product of the Maillard reaction. Its presence in bakery products is one of the most complex challenges within the baking industry. Therefore, its reduction is crucial and represents a key challenge, as special care must be taken not to compromise the characteristics and quality of the product, such as the distinctive colour and flavour of bread. In addition, considering that bakery products are among the most widely consumed foods globally, the levels of acrylamide formed during the baking process inevitably led to exposure to acrylamide through consumption. To this end, the European Union has defined different maximum levels of acrylamide for each food category. Considering the lack of data in the literature concerning acrylamide mitigation in homemade bread, the objective of this study was to evaluate the effects of two concentrations of L-asparaginase (150 and 300 U/kg flour). The obtained results show that fermentation already reduces the levels of L-asparagine. In bread subjected to enzymatic treatment (150 and 300 U/kg flour), the acrylamide content was reduced by 61% and 78%, respectively. However, only the most severe enzymatic treatment reduced the acrylamide content in homemade bread to below the reference limit of 50 μg/kg of **Reg. (EU) 2017/2158**. In conclusion, the use of L-asparaginase as an acrylamide-reducing agent in bread offers a promising prospect for mitigating the health risks associated with the consumption of foods high in acrylamide. However, further research is needed to fully evaluate the impact of this technology on the nutritional and organoleptic properties of bread, as well as to optimize production conditions to maximize the benefits of acrylamide reduction without compromising product quality. Ultimately, the integration of L-asparaginase into bread production represents an important step towards creating safer and healthier foods for consumers worldwide.

CRediT authorship contribution statement

Martina Calabrese: Writing – original draft, Formal analysis. **Lucia De Luca:** Writing – review & editing, Validation, Supervision, Methodology, Conceptualization. **Giulia Basile:** Software, Investigation, Data curation. **Giovanni Sorrentino:** Formal analysis, Data curation. **Mariarca Esposito:** Investigation, Formal analysis. **Fabiana Pizzolongo:** Investigation. **Giovanna Verde:** Formal analysis, Data curation. **Raffaele Romano:** Supervision, Project administration, Methodology, Funding acquisition, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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