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Non-enzymatic browning reaction of isomaltulose

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Abstract

Isomaltulose is a naturally occurring reducing disaccharide composed of glucose and fructose monomers. It is gaining interest as an alternative sweetener to sucrose in past years. It has been successfully used in various food products like chocolate, breakfast cereals, chewing gums, and dairy products especially, ice cream and yogurt. Non-enzymatic browning reactions in foods continue to be an active area of research because of their important roles in color, flavor and nutritional quality. The present study aimed to investigate the influence of different environmental factors on the non-enzymatic browning reaction of isomaltulose. The formation of colored products as a result of the non-enzymatic browning reaction was monitored. The most significant color change occurs in isomaltulose solution under the studied conditions (pH, temperature and amino component), which is a clear indication of a more intense course of the non-enzymatic browning reaction. The most intense staining was observed at pH 10, t = 80°C with the amino component glycine. It has been found that while glucose solutions are coloured more intensely in the presence of lysine, isomaltulose solutions are coloured more intensely in the presence of glycine. In addition to the type of amino component, the degree of staining is influenced by the pH value and the temperature. The results obtained showed that the use of isomaltulose in the composition of foods, that are subject to heat treatment, will affect the color formation of the final product.

1. Introduction

Carbohydrates are the most common constituents in foods (both as natural components and as added ingredients). Carbohydrates in food are important not only as energy sources but also as ingredients that impart texture, colour, and flavor to the final product, as well as components that contribute certain benefits to human health (BeMiller, 2019). A significant part of carbohydrates is used as the main raw material in the production of foods because of their pleasant sweet taste.

Amongst carbohydrates, sucrose (α-D-glucopyranosyl β-D-fructofuranoside) better known as "sugar", has the largest quantitative production and application worldwide (Spillane, 2006). It has been found that excessive consumption of food high in sucrose is one of the reasons for the so-called "diseases of civilization" (Rusu, 2009; Hu and Malik, 2010). For this reason, the interest of producers, nutritionists and consumers in sucrose alternatives is explicable. In this case, isomaltulose is a carbohydrate (isomer of sucrose) that has gained popularity in recent years as a substitute for sucrose. Isomaltulose (6-O-α-d-glucopyranosyl-d-fructofuranose) is a reducing disaccharide that occurs naturally in honey and sugar cane juice (Bárez *et al*., 2000). It is composed of glucose and fructose linked with α-1,6 glycosidic bond instead of α-1,2 as it is the bond in sucrose molecule (Lina *et al*., 2002). Isomaltulose is less hygroscopic, less soluble and more stable to acidity than sucrose (Shyam *et al*., 2018). Commercial isomaltulose is produced from sucrose by enzymatic rearrangement. It is known under the trade name Palatinose™. Hydrogenation of isomaltulose produces isomaltitol (commonly called isomalt) a polyol that is also widely used in the production of some sweet-tasting foods. Isomaltulose has about 50% less pronounced sweet taste compared to sucrose and a relatively low glycemic index GI = 32 (Holub *et al*., 2010; Sawale *et al.,* 2017; Maresch *et al.,* 2017; Shyam *et al*., 2018). Several scientific studies have reported that isomaltulose does not cause tooth decay (Sawale *et al*., 2017; Maresch *et al*., 2017; Shyam *et al*., 2018).

Isomaltulose is “generally recognized as

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safe" (GRAS) by the United States Food and Drug Administration (FDA). In the European Union, it was approved as Novel Food in 2005 (FDA 2006; EU Commission, 2007).

Currently, isomaltulose is used as an alternative to other sugars and maltodextrins in foods and beverages, including sports beverages, energy drinks, malt beverages, foods for special and clinical nutrition feeds, breakfast cereals, cereal bars, dairy products, bakery and pastries, icings, sugar confectionery (e.g., chocolates, jellies, ice cream, chewing gum) and others (Sawale *et al*., 2017; Shyam *et al*., 2018).

Many different chemical reactions take place during the heating and storage of foods, giving rise to the formation of new compounds that affect the final characteristics of foodstuffs and consumer acceptability (Rufián-Henares and Pastoriza, 2016). Some of these transformations include a complex set of reactions known as non-enzymatic browning reactions (NEBR). There are three main mechanisms by which non-enzymatic browning occurs in foods: caramelization, ascorbic acid oxidation and Maillard reactions (Pastoriza *et al.,* 2018). Maillard reactions are used for a group of chemical reactions, initiated by condensation of an amino group with reducing sugar and then followed by a cascade of reactions in foods leading to the formation of different intermediates including aroma components and high molecular weight brown polymers. They were first discovered in 1912 (Maillard, 1912) and the main pathways of their chemistry were described by Hodge in 1953. The complexity of this process lies in the fact that its course depends on a number of factors such as the concentration of reagents (carbonyl and amino component), water activity, temperature, pH and other parameters (Finot *et al*., 1990; Ames, 1992; Laroque *et al*., 2008). The most suitable precursors of the carbonyl components are fructose and glucose. The reaction is not limited to monosaccharides. It also occurs with the reducing disaccharides maltose and lactose (Obretenov, 1983). Among amino components, the most significant precursors are primary amino groups and ammonia (Troise, 2018). The opinion about the participation of glycine, alanine and leucine is contradictory. Some authors deny and others confirm the high activity of these amino acids in the sugar-amine reaction (Obretenov, 1983).

The nature of chemical reactions and the products obtained as a result of the Maillard reaction continue to be studied and discussed to this day, especially when it comes to complex systems such as food (Bhesh *et al*., 2013). For these reasons, the present study aims to investigate the influence of different factors such as pH, temperature and type of amino components on the non-enzymatic browning reaction of isomaltulose.

2. Materials and methods

2.1 Materials

To carry out the study, Isomaltulose under the trade name Palatinose™-Beneo was used, isomalt under the trade name C * IsoMaltidex 16502 (Cargill), fructose from Galam, and glucose - C☆Dex™, (Cargill). 0.1 M glycine (Sigma-Aldrich) and 0.1 M lysine (Sigma-Aldrich) were used as amino components.

2.2 Methods

To evaluate the possibilities for the Maillard reaction, the formation of coloured products in 20% aqueous solutions of isomaltulose, fructose, glucose and isomalt was monitored. For this study, 8 model systems were analyzed: isomaltulose-glycine, isomaltulose-lysine, fructose-glycine, fructose-lysine, glucose-glycine, fructose-lysine, isomalt-glycine, isomalt-lysine. The concentration of sucrose, glucose, fructose and sweeteners were chosen in accordance with often used concentrations of sucrose in most confectionery and pastry products. 0.1M concentration of amino components was used previously in studies regarding the kinetics of the Mayard Browning reaction (Baiser and Labuza, 1992; Davies and Labuza, 1997).

The course of the reaction was considered at pH 9 and pH 10 and temperatures (t) 70°C and 80°C. The formation of coloured products as a result of a non-enzymatic browning reaction was estimated. The samples' colour was determined spectrophotometrically (Carl-Zeiss spectrophotometer, Germany) by measuring the light absorption of the samples at A = 420 nm every 10 min to 100 min (BeMiller, 2010).

In order to evaluate the actual staining of the model solutions of isomaltulose and other reagents, as a result of non-enzymatic browning, controls were also analyzed, in the composition of which the amino component did not participate, analyzed under identical conditions (pH, temperature and duration).

3. Results and discussion

3.1 Alkaline degradation

Figure 1 illustrate the course of alkaline degradation of samples and used as controls without amino acid components. A pH of 9 and temperatures of 70°C and 80°C did not lead to the initiation of alkaline degradation reaction in samples. At pH 10, a change in the color of isomaltulose, glucose and fructose modal solutions was observed at both temperatures. The largest change in color was measured in the model solution of

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isomaltulose, which reaches A_{420} = 1.3. A significant change in colour is also observed in the fructose model solution. With the increase of temperature from 70°C to 80°C a large acceleration in the alkaline degradation reaction of glucose was observed which was proved by the formation of 95% more intensive color (Finot *et al*., 1990). No change in the colour of the isomalt samples was observed. This confirms the fact that polyols do not participate in degradation processes in an alkaline environment (Grembecka, 2015).

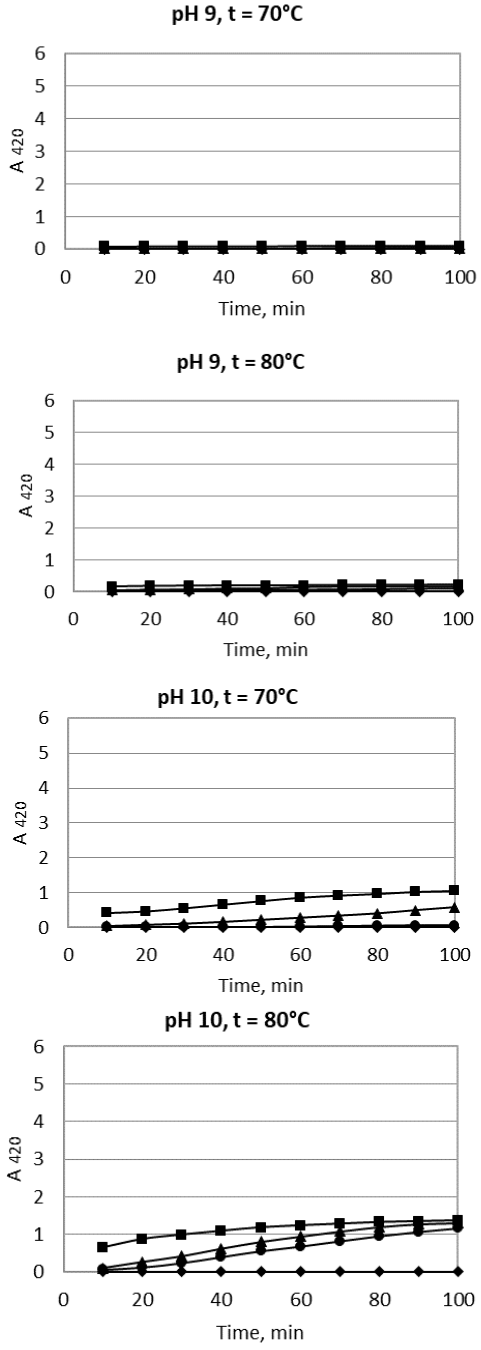

Figure 1. Alkaline degradation, type of sample: ■-isomaltulose, ▲- fructose, ●-glucose, ♦-isomalt

3.2 Non-enzymatic browning reaction

Non-enzymatic browning reaction (Mallard reaction) of isomaltulose, isomalt, glucose, and fructose samples have been evaluated by using glycine and lysine as amino components in order to initiate the reaction.

Figures 2 illustrates the measures of colour change in model solutions with lysine. Evaluation of the non-enzymatic browning reaction of samples with glycine is presented in Figure 3.

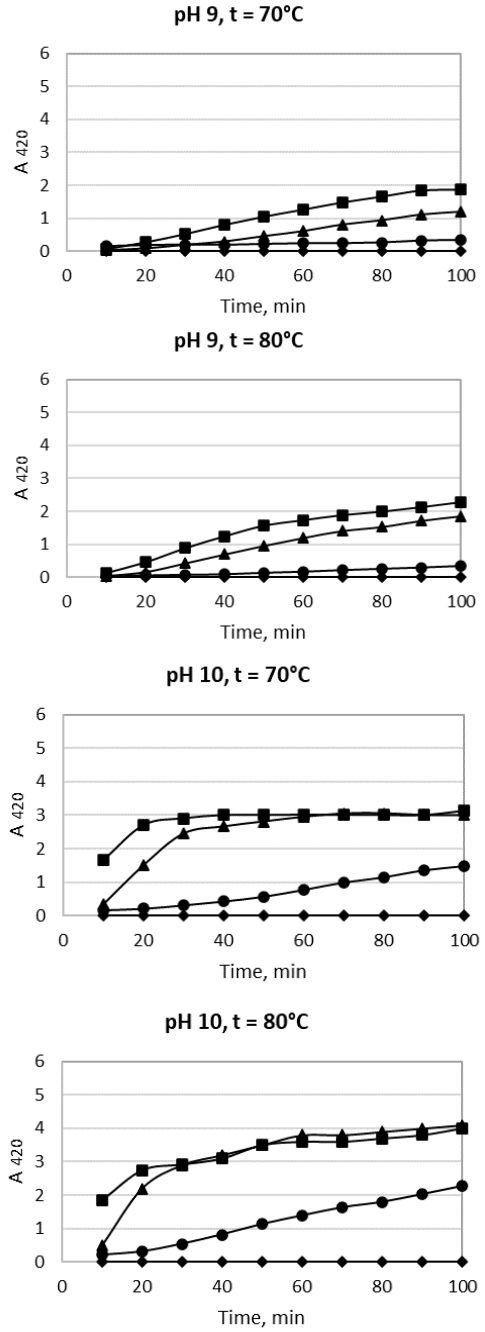

Figure 2. Non-enzymatic browning reaction with the amino component – lysine, type of sample: ■-isomaltulose, ▲-fructose, ●-glucose, ♦-isomalt

Regardless of the pH values, temperature values, and the type of amino component, the most significant change in colour occurred in isomaltulose solution which is a clear indication of more intensive course development of the sugar-amine reaction. A significant positive change in the color progress of the sample was observed at pH 10, t = 80°C with glycine (Figure 3d), where the value of A_{420} reached 5.0 for a retention time of 100 minutes. It was found that at pH 10 and

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temperature 70°C, regardless of the type of amino acids, the colour of the model solutions of isomaltulose changed (reached its maximum value in the 40th-minute retention), after which the intensity of the colour did not change significantly.

From the analyzed data of Figure 2 and Figure 3 it has been confirmed that in addition to the structure of carbohydrates, the type of amino acids also influences the course of the Maillard reaction. While model solutions of glucose stained more intensely in the presence of lysine, model solutions of isomaltulose stained more intensely in the presence of glycine. For example, the solution of isomaltulose at pH 9, t = 70°C per 100 minutes with lysine A_{420} = 1.052 and of glucose A_{420} = 0.340 and of the amino component, glycine isomaltulose is A_{420} = 2.160 while the model solution of glucose is with A_{420} = 0.036.

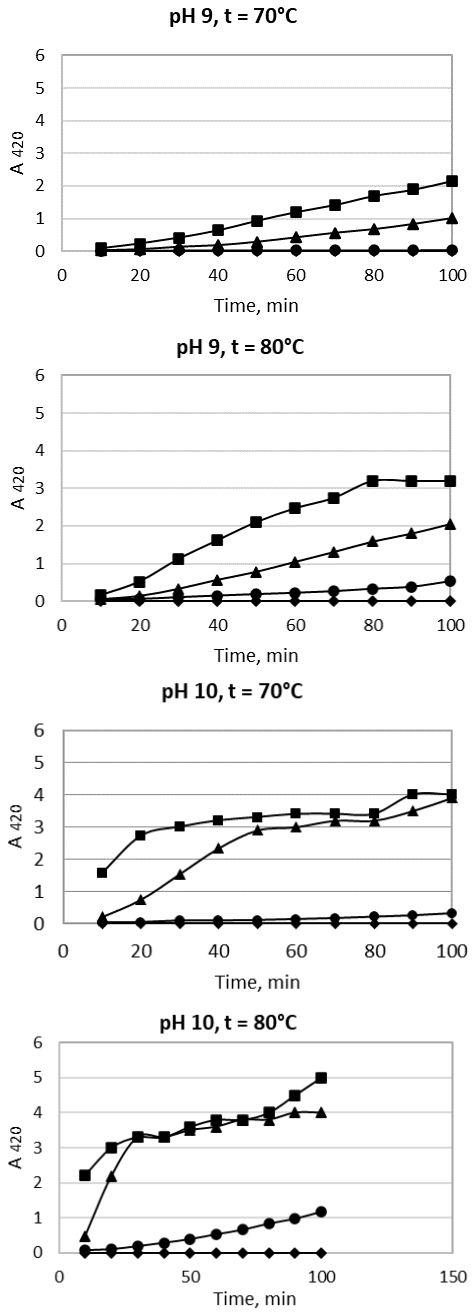

Figure 3. Non-enzymatic browning reaction with the amino component – glycine, type of sample: ■-isomaltulose, ▲-fructose, ●-glucose, ♦-isomalt

In addition to the type of amino components, the degree of staining was influenced by the value of pH and temperature. With the increase in temperature and pH the staining of model solutions was raised, which was confirmed by other studies (Kim and Lee, 2008; Wang *et al.,* 2011). For example, at t = 80°C and pH 9 at 60th minutes retention, the model solution of isomaltulose (with glycine) is about 50% more intensively coloured than its solution at t = 70°C and the model solution of fructose is about 65%. Similar dependencies have been observed when the pH value increases. For example, t = 70°C and pH 9, the light absorption of the isomaltulose model solution prepared with lysine reached 1.266 and that of fructose 0.605. With the rise of pH value to 10 the light absorption of the isomaltulose sample was 3.012 and that of fructose 2.952. A similar relationship was observed in glucose staining.

Regardless of the conditions studied, the color of the isomalt model solution remained unchanged which confirmed that it did not participate in the sugar-amine reaction.

4. Conclusion

From implemented comparative analysis it was found that isomaltulose is actively involved in a non-enzymatic browning reaction. Under the conditions studied (pH, temperature and amino component), its model solution is stained to the highest degree, which is a clear indication of an intense reaction. Staining is highest at pH 10 and temperature 80°C with the amino component glycine. From the obtained results it can be concluded that the use of isomaltulose in the composition of foods that are subject to heat treatment will affect the colour formation of the final product. It has been found that increasing the temperature and the pH value favor the course of the Maillard reaction. It affects both the type of carbohydrate component and the amino acid.

Conflict of interest

The authors declare that they have no conflict of interest.

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