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Chemical Isomerization of Whey Lactose to Lactulose by Using Batch Reaction

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Abstract: The present study was conducted to evaluate the synthesis of lactulose from whey lactose by using 1, 2 and 3M sodium aluminates as alkaline catalyst in batch reaction system at 30 and 70°C. The effect of different sodium aluminates molarities on the isomerization of lactose to lactulose and the time periods were also investigated. The maximum conversion of lactose to lactulose and the by-products formation have been investigated. The HPLC technique was used to determine whey lactulose, B vitamins and the by-products formation. After 1 h at 70°C, by using 3M of sodium aluminates, the isomerization yield was more than 66%. The present study describes an appropriate method for obtaining sweet whey with acceptable yields of lactulose.

Key words: Whey lactose, batch isomerization, lactulose

INTRODUCTION

The disaccharide lactulose is an artificial sugar with a significant impact on human digestion. It can be generated by either alkaline isomerization of lactose via the Lobry de Bruyn-Alberda van Ekenstein rearrangement or by enzyme-catalyzed synthesis. Based on the first reaction, different process schemes for the preparation of lactulose have been developed. The enzymatic synthesis of lactulose can be carried out using different pathways with the transgalactosylation reaction being the most promising (Buhring *et al*., 2010). Lactulose is recognized as a prebiotic and it is used in the form of syrup for the treatment of some intestinal disorders. It is slightly sweeter than lactose and can be used as a partial sucrose substitute in some food products (Drakoularakou *et al*., 2011).

Lactulose can also be used as a food supplement in pediatric diets for the development of functional foods and in geriatric medicine for some targeted populations with severe constipation syndrome (Curiale *et al*., 2013). Lactulose is also widely used as a statement in hepatic encephalopathy.

Lactose is an expensive raw material for lactulose synthesis. Applying milk whey, which contains 4.5-5% (w/v) lactose, is much more cost effective. Whey is a by-product of cheese and casein production and an inexpensive raw material containing a high amount of lactose. Much of the whey produced by the dairy industry is used as animal feed, fertilizer and as a food ingredient or additive (Kassa *et al*., 2008). Nevertheless, about 30% of the annual world whey production (177 million tons in 2006) remains underutilized (Alonso *et al*., 2011). Whey is considered an environmentally high-strength wastewater pollutant due to its high biological oxygen demand and chemical oxygen demand (Abboud *et al*., 2010). Its cost-effective disposal and utilization has become increasingly important to the dairy industry. Additionally, environmental and economic concerns demand that whey should be converted to value-added products.

The aim of the present study was to study the effect of different molarity of sodium aluminates, temperature degree (70°C) and sweet whey lactose for different periods of time on the formation of lactulose.

MATERIALS AND METHODS

Aluminum wires of 99.9% purity were used to prepare sodium aluminate. Aluminum wires were cut into 3 to 5 mm length pieces and dissolved in sodium hydroxide solution. Fifty four gram of cutting aluminum wires were mixed with 1L of 2M sodium hydroxide in 2L beaker. A hot plate magnetic stirrer was used to perform the reaction in a hood fitted with vacuum unit. The reaction is an exothermic, so we did not need to use heating. After dissolving all of aluminum cuttings, the mixture was allowed to cool to room temperature. The mixture was filtered through Whitman paper No. 54. The precipitated sodium aluminate was washed with distilled water to get rid of access base.

The sweet whey in the present work was obtained from the Food Science Department Teaching Plant in the College of Agriculture, University of Baghdad. The whey was heated 15 min. at boiling point to separate the whey proteins as a curd. The proteins free whey was filtered by using Buchner funnel through Whitman filter paper No. 1, to get rid of whey protein residues and getting a clear solution. The obtained solution was stored under refrigeration in a sealed container for the subsequent analysis and isomerization processes (Dominguez, 1984).

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500 mL of sodium aluminates was used. The isomerization was carried out at temperature 30 and 70°C for various periods (1, 2, 3, 4, 5, 6 and 7 h) with 4.2% sweet whey lactose. At the end of each time, a 50 mL of mixture was taken and the pH was adjusted to 7 by using 10% phosphoric acid. The precipitation Aluminum phosphate was separated by using centrifuge at 4,000 rpm for 10 min the supernatant was collected and stored at refrigeration. High performance liquid chromatography (HPLC) method was used to determine the degree of isomerization of lactose into lactulose, following the procedure of Cataldi *et al.* (2000). Anion exchange shimpack A1 column, 3 µm particle size, with a column diminution of (50 cm x 4.6 mm). The mobile phase was a mixture of 15 mM NaOH spiked with 1mM barium acetate which was prepared freshly 1.5 mL/min. flow rate. The detector was Refractive index. The column temperature was controlled at 25°C. The injection size was 20 µL.

HPLC method was used to determine sweet whey lactose and B group vitamins.

RESULTS AND DISCUSSION

Whey is a by-product that has been traditionally considered as a residue of cheese. The magnitude of the dairy industry makes whey production enormous volumes and it becomes a very large organic pollutant for the environmental. Few large companies in the world have the capacity to implement different technologies for the efficient recovery and upgrading of whey (Constanta, 2012). The chemical composition of whey is dependent upon chemical composition varies according to slight changes in milk processing parameters (Polat, 2009). Cheese whey represents about 90-95% of the milk volume and retains about 55% of milk nutrients. The most abundant of these in sweet whey are lactose and B group vitamins (Marwaha and Kennedy, 1988).

Before the isomerization processes the sweet whey was subjected to lactose content and vitamins B group determinations. HPLC was used to determine whey lactose and B group vitamins. It was found that the lactose concentration of purified free-protein sweet whey (which is used in batch reaction) was 4.2% w/v.

As shown in Fig. 1, there was a direct proportional relationship between lactulose formation in the sweet whey and temperature by using 1M sodium aluminates as alkaline catalyst in the isomerization reaction. Similar, relation was found between lactulose formation and the period of reaction. The direct proportional relationship was true till 4 h of isomerization reaction, but there was a gradual decreasing in lactulose percentages over the 4 h reaction. In temperature degrees (30 and 70°C) of isomerization of sweet whey lactose to lactulose by using 1M sodium aluminates showed a high lactulose formation during the 1st h of reaction. After one hour of the isomerization, the percentage of lactulose formation were 62.50, 63.63% (of the maximum amount of lactulose after 4 h) at 30 and 70°C, respectively.

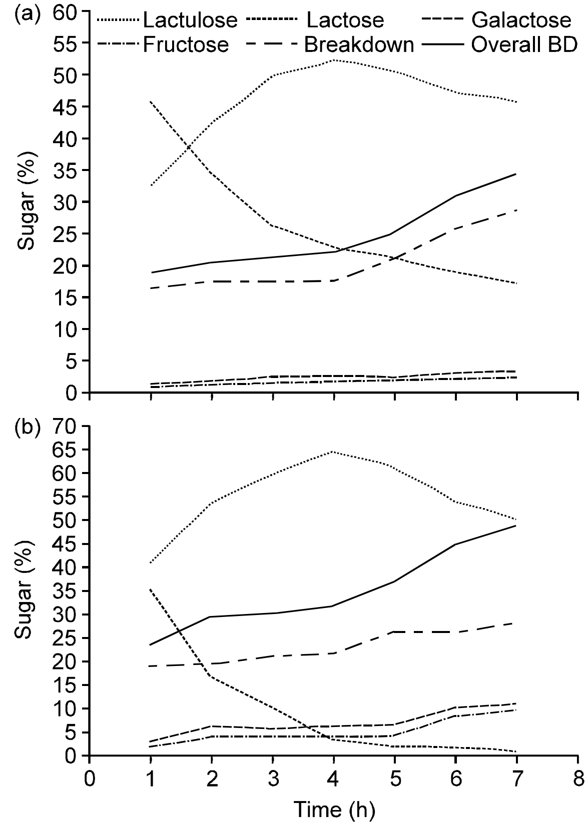

Fig. 1: Effect of 1M sodium aluminates on the formation of lactulose by using sweet whey of 4.2% lactose at: (A) 30°C and (B) 70°C

Fig. 1 shows that the overall degradation also had a direct proportional relationship with temperature degrees, the overall degradation was more during the isomerization process of sweet whey lactose by 1M sodium aluminates. In addition, the overall degradation at the end of the isomerization (in this study after 7 h), by using 1M sodium aluminates was more than using 2M and 3M sodium aluminates. Fig. 1 shows that there was a direct proportional relationship between the formation of galactose and both the temperature and time. A similar relationship was distinguished with the formation of fructose.

Figure 2 shows that the main quantity of lactulose produce during the first hour of isomerization reaction by using 2M sodium aluminates as alkaline catalyst and the increasing was gradually ascending up to the 4th h before it gradually decreased. Upon the 1st h of the isomerization, the percentages of lactulose were 60.60 and 62.11% of the higher percentages (after 4 h) of

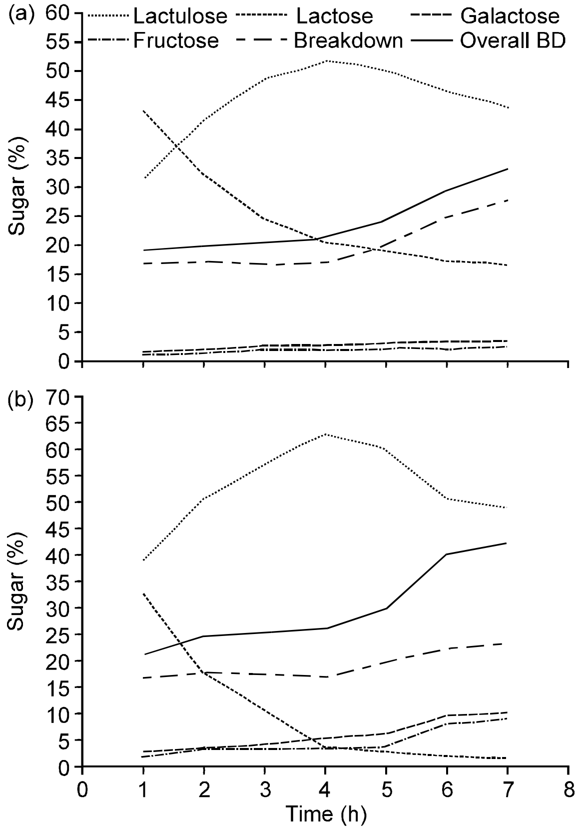

Fig. 2: Effect of sodium aluminates concentration 2M on the formation of lactulose by using sweet whey 4.2% lactose at: (A) 30°C and (B) 70°C

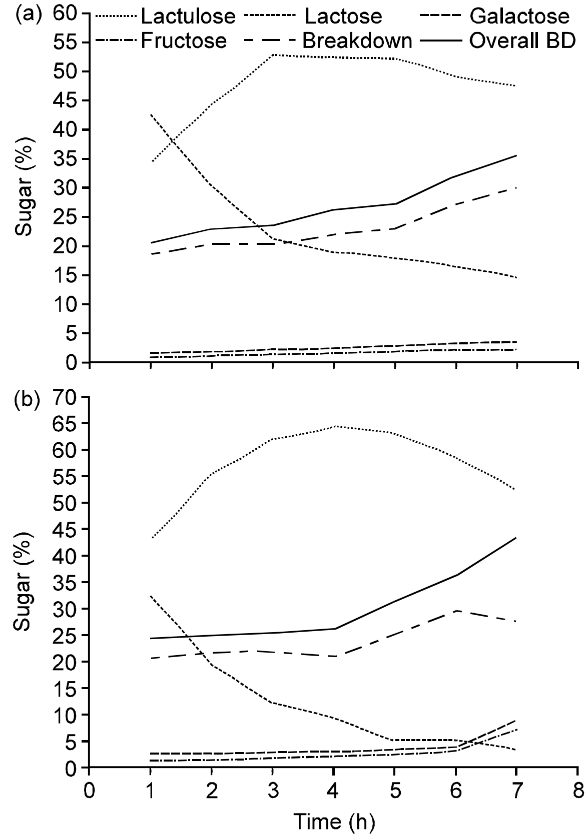

Fig. 3: Effect of sodium aluminates concentration 3M on the formation of lactulose by using sweet whey of 4.2% lactose at 70°C

lactulose at 30 and 70°C. If a comparison was made between the amounts of lactulose produced during the first hour at different temperature degrees, we can find that the percentage of lactulose increased by 17.61% at 70°C as compared with 30°C.

Figure 2 shows that the overall degradation was increased after 7 hr. of the isomerization by 22.48% 70°C as compared with 30°C.

As compared with 1M, 2M sodium aluminates, economically, gave insignificant increasing in lactulose formation especially after 1 h of reaction. Increasing in the first hour by using 2M sodium aluminates at 30 and 70°C were more by 2.89 and 2.00% than these percentage obtained by using 1M sodium aluminates, this result agreed with Zokaee *et al*. (2002) who showed the increase in molar ratio of aluminates to lactose from 1 to 2, the rate of reaction and maximum conversion were increased.

The lactulose formation averages in sweet whey have been increased by using 3M sodium aluminates as compared with 1 and 2M. The initial formation stages of the overall degradation in sweet whey were also higher as compared with 1 and 2M.

Figure 3 shows that there was a direct proportional relationship between the percentage of lactulose formation and the time of reaction. A similar relationship was found between the overall degradation formation and time. Again, the main quantity of sweet whey lactulose was formed during the first hour of the isomerization 65.62 and 66.31% of the higher quantity of lactulose (after 4 h) was formed during one hour of isomerization reaction. A direct proportional relationship was found between galactose and fructose formations with both temperature and time, this result agreed with Buhring *et al*. (2010) who showed the initial time of reaction giving the main percentage of lactulose maximum quantity, that because there was not enough lactulose on the other side of the chemical equation to oppose the formation of more lactulose, as soon as the lactulose was accumulated with the by-products, the averages of lactulose formation going to be reduced. The by-products formation always accompanied high molarity alkaline and high temperature degree.

The overall degradation as we mentioned before, is a mixture of nutritives sugars (some were prebiotic such as tagatose) (Bertelsen *et al*., 1999; Schumann, 2002); that's mean the sweet whey drinks would be enriched many kinds of sugars were compared with lactose only.

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