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Optimization of Sucrose Substitution in Lassi by Tagatose as a Sweetener

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A B S T R A C T

Reduction in consumption of sugar is becoming need of the hour, due to its adverse effects on health. Use of alternative low calorie sweeteners have several limitations in their applications. Tagatose appearing as highly promising low calorie sweetener for food applications. In present study suitability of tagatose was evaluated as a sweetener in Lassi. Findings of the study suggest that in preparation of Lassi, from 12 g sucrose 9 g (75%) can be substituted by using 10.8 tagatose along with 3.0 g sucrose per 100 ml. Similarly, in preparation of Lassi, 12 g sucrose can be completely substituted by using blend comprising of 12.96 tagatose and 1.44g fructose as a sweetener per 100 ml.

Introduction

Sucrose is gold standard for sweetness and most widely used sweetener, but it contributes excess calorie and causes overweight and obesity, which in turn leads to several health issues including type 2 diabetes, heart attack, kidney disease, some cancers and tooth decay (Levin, 2002; Oliveira *et al.*, 2015; Van Laar *et al.*, 2021). The World Health Organization recommends that added sugars should make up no more than 10 per cent of daily caloric intake, with a proposal to lower this level to 5 per cent or less for optimal health (Mooradian *et al.*, 2017). Therefore, reduction in sucrose intake becomes necessary, but none of the alternative sweetener found ideal (Luo *et al.*, 2019; Mariotti & Lucisano, 2014). Fortunately, now tagatose is emerging as a promising sucrose substitute due to its relatively high sweetness, low caloric content, low glycemic index, desirable physicochemical properties for food application with several health benefits (Rouhi *et al.*, 2015).. In India, among sweetened fermented dairy products, Lassi is the most popular and widely consumed traditional Indian dairy product all throughout the

country. Therefore, from the category of fermented milk products Lassi was selected to try substitution of sucrose by tagatose as a sweetener.

According to Fujimaru *et al.,* (2012), in practice the sensory profile of sweeteners may be affected by factors such as temperature and characteristics of food systems in which the sweeteners are used. It appeared from this report that equivalent sweetness of tagatose may vary in different types of dairy products, as serving temperature and characteristics of different types of dairy products vary widely. Therefore, it is necessary to ascertain effect of replacement of sucrose by tagatose on acceptability of dairy products belonging to different categories and optimize the use of tagatose as substitute of sucrose. The work was carried out for products of different categories and results of Lassi belonging to fermented dairy products are presented here.

Materials and Methods

The tagatose of Nu Natural brand was procured from local supplier of ingredients. The content (purity) of tagatose was 100 per cent. Sucrose of Madhur brand from "Shree Renuka Sugars Limited" was procured from local market. Content (purity) of sucrose was 99.4 per cent. Fructose of "Lobachemie" brand was purchased from local supplier. Content (purity) of fructose was 99 per cent. Microbial culture: Cultures Streptococcus thermophilus (MTCC 5460) and *Lactobacillus helveticus* (MTCC 5463) were obtained from Dairy Microbiology Department of this college.

Preparation of Lassi

For preparation of Lassi, Amul brand toned milk containing 3 per cent fat and 8.5 per cent SNF was used for the preparation of Lassi. The Lassi available in Indian market usually contains 12 per cent sucrose (Aneja *et al.,* 2002; De, 2001). Therefore, in this study for preparation of Lassi sucrose was added at rate of 12 per cent.

In industrial production of Lassi, addition of sucrose is reported at the rate of 12 per cent (Ranganadham, 2016). Therefore, in this study for preparation of Lassi sucrose was added at rate of 12 per cent.

Lassi was prepared using method as reported by Aneja *et al.,* (2002). The milk was heated to 90°C for 10 minutes. After cooling milk to 40°C mixed culture (*Streptococcus thermophilus*(MTCC 5460) and *Lactobacillus helveticus*(MTCC 5463), in ratio of 1:1) was inoculated at the rate of 1 per cent and incubated for 12 hours at 42°C.

After incubation sweetener and water was added to give sweetens and reduce viscosity. Product was stirred to smooth uniform consistency. Addition of water was kept fixed at 20 per cent by weight of curd. The amount of sweetener was varied as per the details given in Table 1.

All the samples of Lassi were filled in PET bottles and stored in refrigerator for overnight at 6±1°C. The samples well mixed before offered to the panel of judges for evaluation of their overall acceptability in sensory evaluation using 9 point hedonic scale.

Sensory evaluation of Lassi

Sensory evaluation of flavoured milk, Lassi, ice-candy, burfi and sandesh was carried out for their overall acceptability using 9 point hedonic scale according to the method suggested by Wichchukit & O'Mahony (2015). The evaluation was carried out by a panel of trained judges. The score given by different judges was averaged out. The average values were compiled and used for statistical analysis and reported here.

Statistical analysis of data

The mean values of each of the attributes in the study were subjected to statistical analysis using Completely Randomized Design. The statistical model adopted was given by Steel and Torrie (1980) which is illustrated as given below:

$Yij = m + Ti + Eij$

Where,

Yij = Response due to j^{th} observation in the i^{th} treatment.

m = general mean

Ti = effect of i^{th} treatment, and

Eij = Error due to j^{th} observation in the i^{th} treatment.

Results and Discussion

For substitution of sucrose by tagatose as a sweetener in the dairy products, initially three different possibilities were explored, as listed below.

Substitution using tagatose alone as sole sweetener for complete (100%) substitution of sucrose

Substitution using blend of tagatose and sucrose as sweetener for partial (25, 50 and 75%) replacement of sucrose

Substitution using blend of tagatose and fructose as sweetener for complete (100%) replacement of sucrose

It is well known that in acceptance or rejection of food products appearance, color, odor, taste and texture are primary determinants (Izutsu *et al.,* 1981). Therefore, sensory evaluation is a key point in market success of food products. Taste perception plays a vital role in the sensory evaluation process (Zargaraan *et al.,* 2016). Hence, all the samples of Lassi prepared in the study were evaluated for their overall acceptability in sensory evaluation using 9 point hedonic scale. The panelists of the judging committee were requested to consider taste and aroma, colour and appearance as well as mouth feel of the Lassi in deciding its overall acceptability score.

Complete substitution sucrose using tagatose alone as sole sweetener

Since bulk properties of sucrose and tagatose are reported to be similar to that of the sucrose, possibilities of straight forward complete substitution of sucrose by using in different amount of tagatose were explored in the first approach. Keeping in view the reported relative sweetness of tagatose as 0.9 (*i.e*. 90%) and sour taste of Lassi, the rate of tagatose addition was kept at the rate of 0, 10, 20 and 30, per cent higher than that of the addition of sucrose. Accordingly, samples of Lassi were prepared using tagatose at the rate of 12.0, 13.2, 14.4 and 15.6 g per 100 ml. The Lassi with 12.0 per cent sucrose was also included to serve as control sample in comparison with the sucrose substituted samples of Lassi. The average of results obtained from four replications are presented in Table 2.

It was evident from the results that the overall acceptability score of Lassi increased upon increasing the amount of tagatose from 12.0 g per 100 ml to 14.4 g per 100 ml. However, further increase in rate of tagatose addition to 15.6 g per 100 ml, there was noticeable decrease in the overall acceptability score of the Lassi, giving lowest score among all the samples under comparison. The highest overall acceptability score of was obtained when the Lassi was prepared using 12 per cent

sucrose as a sweetener. Statistical analysis of the data revealed that the substitution of sucrose by tagatose as a sweetener has no statistically significant effect on overall acceptability score of the Lassi. The overall acceptability score of all the samples of Lassi prepared using 12.0 to 15.6 g tagatose per 100 ml was statistically at par with each other. Similarly, the overall acceptability score of all the samples of Lassi prepared using 12.0 to 15.6 g tagatose per 100 ml was statistically at par with that of the Lassi prepared using 12.0 sucrose per 100 ml.

It became very clear from the above results that the overall acceptability score of all the samples of Lassi prepared using different level of tagatose was at highly acceptable level and almost equivalent to that obtained for Lassi prepared using 12 g sucrose per 100 ml. The use of tagatose in Lassi at the rate of 14.4 g per 100 ml gave the overall acceptability score almost equivalent to that obtained for Lassi prepared using 12 g sucrose per 100 ml. Thus, addition of tagatose in amount 20 per cent higher than sucrose gave the best results for acceptability in sensory evaluation. Therefore, addition of tagatose as a sole sweetener in preparation of Lassi at the rate of 14.4 g per 100 ml was selected for further study.

These results suggested that the use of tagatose alone as a sole sweetener was capable to produce the highly acceptable Lassi. Hence, tagatose alone may be used for complete substitution of sucrose in preparation of Lassi. Hence, for complete substitution of sucrose in Lassi by tagatose alone as a sole sweetener, from different amounts of tagatose tried, the use of tagatose at the rate of 14.4 g per 100 ml was selected for further work. The decrease in the overall acceptability score of Lassi up on addition tagatose at the rate of 15.6 g per 100 ml might be attributed excessive total solids content of the product, compared to normal level.

The encouraging results obtained in Lassi were highly surprising compared to those obtained in case of the flavoured milk. The logical reasons for significant difference in performance of tagatose as a sweetener in Lassi and that in flavoured milk lower overall acceptability of the Lassi prepared using tagatose as a sweetener, might be attributed to medium. Flavoured milk is near neutral product, whereas, Lassi is highly acidic product.

It is reported that the taste quality and intensity are dependent not only on a compound's structure but on other taste reception parameters: temperature, pH, viscosity and the presence of additional sweet or non-sweet compounds (Belitz *et al.,* 2009). It is also pointed out by Fujimaru *et al.,* (2012) that in practice the sensory profile of sweeteners may be affected by factors such as temperature and characteristics of food systems in which the sweeteners are used. These views were further substantiated from study conducted by Paixão*et al.,* (2014). These authors demonstrates that the food matrix and product consumption temperature can dramatically influence the behavior of sweetening compounds in an unpredictable way. Therefore, findings of present study may be used by researchers and dairy processors while developing milk based beverages.

It appeared from the literature that no work is reported so far regarding use of tagatose as a sweetener in the Lassi, as prepared in present study. Therefore, results of the present study could not be compared as such with the literature. However, in soft drinks it has been found that tagatose shows significant synergistic effects at even low doses with combinations of intense sweeteners. When blended with intense sweeteners, tagatose improves flavor and mouth-feel. It also seems that tagatose is able to stabilize the sweetness and flavor profile in soft drinks sweetened

with blends of aspartame and acesulfame potassium. Adding as little as 0.2 per cent tagatose to the sweetener blend provides an element of stable sweetness, and the tagatose is able to mask the bitter taste of acesulfame potassium, thereby prolonging the shelf-life of the soft drink (Bertelsen *et al.,* 2001). Therefore, somewhat similar phenomenon might be playing role in better performance of tagatose as a sweetener in Lassi.

Partial substitution sucrose using blend of tagatose and sucrose sweetener

As discussed above that the substitution of sucrose by tagatose alone as a sweetener in preparation of Lassi, the complete substitution gave the Lassi having overall acceptability score statistically at par with the Lassi prepared with sucrose alone as a sweetener. However, looking to the surprising results obtained in the first approach, it was felt worth to examine combination of tagatose and sucrose as a sweetener in Lassi for possibility of their synergistic or antagonistic effect. Therefore, in the second approach for substitution of sucrose by tagatose in Lassi, possibilities for partial substitution of sucrose by tagatose was examined.

In partial substitution 3, 6, 9 and 12 g sucrose was substituted by 3.6, 7.2, 10.8 and 14.4 g tagatose per 100 ml to give 25, 50, 75 and 100 % substitution of sucrose. The 100% substitution of sucrose by tagatose was also included here to serve as a control. The Lassi with 12 g sucrose per 100 ml was also included to serve as control sample in comparison with the sucrose substituted samples of Lassi. The average of results obtained from four replications are presented in Table 3.

It was evident from the overall acceptability score of the Lassi increased gradually with increase in rate of sucrose substitution. The overall acceptability score remained highest in case of the Lassi prepared using 75 per cent substitution of sucrose with tagatose.

The lowest overall acceptability score was obtained in case of the Lassi prepared with 25 per cent substitution of sucrose with tagatose.

Though, there were some differences in overall acceptability score of different samples of Lassi, the statistical analysis of the data revealed the differences were statistically non-significant. The overall acceptability score of all samples of Lassi prepared from substitution of sucrose by tagatose were at par with each other. Moreover, the overall acceptability score of all samples of Lassi prepared from substitution of sucrose by tagatose were at par with that of the Lassi prepared from sucrose as a sole sweetener.

Since, use of blend consisting of tagatose and sucrose at the rate of 10.8 g and 3.0 g respectively per 100 ml as a sweetener in Lassi gave the highest overall acceptability score.

Therefore in partial substitution of sucrose in Lassi, among various blends of tagatose and sucrose tried in the study, use of blend consisting of tagatose and sucrose at the rate of 10.8 g and 3.0 g respectively per 100 ml was selected for further study.

The relative better performance of this blend in imparting sweetness in Lassi might be attributed to phenomenon of synergy between these two sugars, when used in the specific proportion in the blend.

It appeared from the literature that no work is reported so far in the literature regarding partial substitution of sucrose by tagatose as a sweetener in preparation of Lassi. Therefore, results of the present study could not be compared with the literature.

Complete substitution of sucrose using blend of tagatose and fructose

Inthe third approach for substitution of sucrose by tagatose in Lassi, possibilities for complete substitution of sucrose by using small amount of suitable adjunct sweetener along with tagatose as a major sweetener were explored. Fructose is commercially used as sweetener in foods and beverages due to its low cost and high relative sweetness (Hanover and White, 1993), Looking to these advantages in use of fructose in food, in the present study fructose was also selected as a sweetener. However, keeping in view possibility of some adverse health effects on consumption of fructose in large amount, in present study use of fructose was limited as an adjunct sweetener only.

For use of fructose as an adjunct sweetener with tagatose, blend of tagatose and fructose were formed in the amount of12.96 + 1.44, 11.52 + 2.88 and 10.08 + 4.32 to replace 10, 20 and 30% tagatose respectively. Samples of Lassi containing 14.4 g tagatose alone and that containing 12 8 g sucrose alone per 100 ml were also included to serve as controls while comparison during sensory evaluation. The average of results obtained from four replications are presented in Table 4.

It was evident from the results that initially overall acceptability score of the Lassi increased gradually with increase on further substitution of tagatose by fructose, when compared with overall acceptability score of the Lassi prepared using tagatose alone as a sweetener. However, on further substitution the overall acceptability score of Lassi decreased. The overall acceptability score remained lowest in case of the Lassi prepared using blend of tagatose and fructose as a sweetener at the rate of 10.08 and 4.32 g respectively per 100 ml. The highest overall acceptability score was obtained in case of the Lassi prepared when sucrose was used as a sole sweetener at the rate of 8 g per 100 ml. It appeared from the results that partial substitution of tagatose by fructose on weight by weight basis had no statistically significant effect on overall acceptability score of the Lassi. The overall acceptability score of the Lassi. Prepared using various blend of tagatose and fructose as a sweetener, remained statistically at par with overall acceptability score of the Lassi prepared using tagatose or sucrose alone as a sweetener.

Thus, use of fructose as an adjunct sweetener with tagatose, in the combination of tagatose and fructose provided relatively better performance in complete replacement of sucrose as a sweetening agent in Lassi.

Therefore, in complete substitution of sucrose in Lassi by tagatose alone along with fructose as an adjunct sweetener, from different blends of tagatose and fructose tried, the blend of tagatose and fructose at the rate of 12.96 g and 1.44 g respectively per 100 ml, giving 100 per cent sucrose substitution was selected for further work.

The improvement obtained in overall acceptability score of Lassi upon use of fructose as an adjunct sweetener with tagatose as major sweetening agent might be attributed to its high relative sweetness (Shallenberger, 1993; Barclay *et al.,* 2012). Another reason might be attributed to its synergistic effect of fructose, because synergistic effect of fructose in improving taste is very well documented in literature (Van Tornout *et al.,* 1985; Schiffman *et al.,* 1995; Barclaya *et al.,* 2012; Reyes *et al.,* 2019).

It appeared from the literature that no work is reported so far in the literature regarding use of fructose as an adjunct sweetener with tagatose in Lassi, Therefore results of the present study could not be compared with the literature.

Table.1 Amount of sweeteners used in preparation of Lassi

Sweetener	Amount used (g/100 ml Lassi)
Tagatose	12.0, 13.2, 14.4 & 15.6
Tagatose + Sucrose	14.4 + 0.0, 10.8 + 3.0, 7.2 + 6.0 & 3.6 + 9.0
Tagatose + Fructose	14.40 + 0.00, 12.96 + 1.44, 11.52 + 2.88 & 10.08 + 4.32
Sucrose	12.0

Table.2 Overall acceptability score of Lassi prepared by complete substitution of sucrose with tagatose

Sr. No.	Sweetener used	Amount added (g/100 ml)	Acceptability score (out of 9)
1.	Tagatose	12.0	8.05[a]
2.	Tagatose	13.2	8.13[a]
3.	Tagatose	14.4	8.36[a]
4.	Tagatose	15.6	7.97[a]
5.	Sucrose	12.0	8.38[a]
ANOVA			
SEm.		0.14	
CD		0.69	
Test$_{(0.05)}$		NS	
CV%		3.87	

Table.3 Overall acceptability score of Lassi prepared by partial substitution of sucrose with tagatose

Sr. No.	Rate of sucrose substitution (%)	Amount of tagatose & sucrose added (g/100 ml)	Acceptability score (out of 9)
1.	100	Tagatose (14.4) + Sucrose (0.0)	8.32[a]
2.	75	Tagatose (10.8) + Sucrose (3.0)	8.35[a]
3.	50	Tagatose (07.2) + Sucrose (6.0)	8.23[a]
4.	25	Tagatose (03.6) + Sucrose (9.0)	8.16[a]
5.	0	Tagatose (00.0) + Sucrose (12.0)	8.28[a]
ANOVA			
SEm.		0.10	
CD		0.49	
Test$_{(0.05)}$		NS	
CV%		2.71	

Table.4 Overall acceptability score of Lassi prepared by complete substitution of sucrose with blend of tagatose and fructose

Sr. No.	Rate of sucrose substitution (%)	Amount of tagatose, fructose &/or sucrose added (g/100 ml)	Acceptability score (out of 9)
1.	100	Tagatose 14.40 + Fructose 0.00	8.21[a]
2.	100	Tagatose 12.96 + Fructose 1.44	8.29[a]
3.	100	Tagatose 11.52 + Fructose 2.88	8.18[a]
4.	100	Tagatose 10.08 + Fructose 4.32	8.14[a]
5.	0	Tagatose 00.00 + Sucrose 12.00	8.36[a]
ANOVA			
SEm.		0.13	
CD		0.64	
Test$_{(0.05)}$		NS	
CV%		3.57	

Fig.1 Lassi prepared using different sweeteners

Table.5 Overall acceptability score of Lassi prepared using different sweeteners

Sr. No.	Rate of sucrose substitution (%)	Sweetener & its amount used (g/100 ml)	Acceptability score (out of 9)
1.	100	Tagatose (14.40)	8.28[a]
2.	75	Tagatose (10.80) + Sucrose (3.00)	8.37[a]
3.	100	Tagatose (12.96) + Fructose (1.44)	8.41[a]
4.	0	Sucrose (12.00)	8.47[a]
ANOVA			
SEm.		0.11	
CD		0.45	
Test$_{(0.05)}$		NS	
CV%		2.53	

Comparison of different approaches for substitution of sucrose by tagatose

After completing evaluation all the different approaches individually for substitution of sucrose in Lassi (straight forward complete substitution, partial substation on equivalent sweetness basis and complete substitution along with adjunct sweetener), work was planned to make comparison between different approaches. The work on comparison between different approaches also aimed to reconfirm the consistency in performance of the promising concentrations of sweeteners selected from different approaches.

In study for comparison of different approaches the samples of Lassi were prepared using 14.4 g tagatose, blend of 10.8 g tagatose + 3.0 g sucrose and blend of 12.96 g tagatose + 1.44 g fructose per 100; corresponding the substitution of sucrose by 100, 76 and 100% respectively. The samples of Lassi were also included using 12.0 g sucrose to serve as a control in the comparison. Photograph of Lassi prepared using (1) tagatose alone, (2) blend of tagatose with sucrose, (3) blend of tagatose with fructose and (4) sucrose alone as sweetener is presented in Figure 4.2.The average of results obtained from four replications are presented in Table 4.

It was evident from the results that the overall acceptability score remained lowest in case of the Lassi prepared using tagatose alone as a sweetener at the rate of 14.4 g per 100 ml. The overall acceptability score of the Lassi was improved slightly upon using of the blend of tagatose and sucrose. The overall acceptability score of the Lassi was further increased slightly upon using of the blend of tagatose and fructose. However, the highest overall acceptability score was obtained in case of the Lassi prepared when sucrose was used as a sole sweetener at the rate of 12.0 g per 100 ml.

It appeared from statistical analysis of data that different approaches used for substitution of sucrose in by tagatose as a sweetener had no significant effect on overall acceptability score of Lassi. The overall acceptability score of Lassi prepared using (a) tagatose alone as a sweetener at the rate of 12 g per 100 ml, (b) blend of 10.8 g tagatose and 3.0 sucrose per 100 ml, (c) blend of 12.96 g tagatose and 1.44 g fructose per 100 ml and (d) sucrose alone as a sweetener at the rate of 12 g per 100 ml was statistically at par with each other. Therefore results of this study clearly remonstrated demonstrated that in preparation of Lassi75 per cent sucrose could be successfully

substituted without significant decrease in its overall acceptability score. Similarly, this study also clearly remonstrated demonstrated that in preparation of Lassi sucrose could be completely substituted without significant decrease in its overall acceptability score, by using blend of tagatose and fructose 12.96 g tagatose and 1.44 g fructose per 100 ml. The results obtained in this comparative study, reconfirmed the highly promising performance of the selected approaches and specific combinations used there in for substitution of sucrose by tagatose in preparation of Lassi.

It appeared from review of literature that no work is reported so far in the literature regarding partial substitution of sucrose by tagatose in Lassi. Similarly, it also appeared from the literature that no work is reported so far in the literature regarding use of fructose as an adjunct sweetener with tagatose in Lassi for partial or complete substitution of sucrose, Therefore results of the present study could not be compared with the literature. Based on findings obtained from entire study on substitution of sucrose by tagatose in preparation of Lassi, the partial (75%) substitution of sucrose by equivalent sweetness base using blend of 10.8 g tagatose and 3.0 g sucrose per 100 ml as well as complete substitution of sucrose by using fructose as an adjunct sweetener along with sucrose in form blend consisting 12.96 g sucrose and 1.44 g tagatose per 100 ml.

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