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Effect of Cyclodextrinase on Dough Rheology and Bread Quality from Rice Flour

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Gluten-free breads are usually characterized by deficient quality characteristics as compared to wheat breads. Problems related to volume and crumb texture are associated with gluten-free breads even when rice flour is used, which seems to be the best raw material for this type of bread. The potential use of cyclodextrin glycosyl transferase (CGTase) as a rice bread improver is presented. The effect of CGTase addition to rice flour on dough rheology and bread quality was investigated. In addition, an experimental design was developed to optimize the levels of CGTase, hydroxypropylmethylcellulose (HPMC), and oil. The addition of CGTase produced a reduction in the dough consistency and also in the elastic modulus. With regard to the rice bread quality, better specific volume, shape index, and crumb texture were obtained. The amount of cyclodextrins in the bread crumb was quantified to explain the action of this enzyme. The data indicate that the improving effect of the CGTase results from a combination of its hydrolyzing and cyclizing activities, the latter being responsible for the release of cyclodextrins, which have the ability to form complexes with lipids and proteins.

KEYWORDS: Rice flour; enzymes; cyclodextrin glycosyl transferase; cyclodextrin; HPMC; dynamic rheology; rice bread quality

INTRODUCTION

Celiac disease, first considered to be a gastrointestinal disease, is a gluten-sensitive entheropathy with genetic, immunologic, and environmental bases. Peptides released from wheat gluten during digestion are responsible for the primary intolerance in genetically predisposed individuals. Although the mechanism is still unclear, cereals containing prolamins such as wheat, rye, barley, and probably oat are toxic for celiac patients, whereas corn and rice are considered to be safe. Later studies suggest an increasing prevalence of celiac disease of up to 1 in 200−300, likely due to the development of new and more sensitive methods of screening (*1*, *2*). The only way to ameliorate the symptoms is keeping the diet of celiac patients as gluten-free as possible (*3*).

The main gluten-free cereals recommended for celiac patients are rice, corn, sorghum, and buckwheat, but with the exception of rice they have negative effects on product quality when used even at concentrations of 10−20% (*4*). Rice flour has many unique attributes such as bland taste, white color, ease of digestion, and hypoallergenic properties. Low levels of protein and sodium, the absence of gliadin, and the presence of easily digested carbohydrates make it an ideal food for patients suffering from celiac disease. However, despite the numerous advantages of rice flour, the lack of gluten proteins makes it very difficult to obtain an acceptable yeast-leavened product such as bread because of the absence of a proper network necessary to hold the carbon dioxide produced during proofing. Different approaches have been presented to overcome that problem; xanthan gum and carboxymethylcellulose (CMC) have been used as gluten substitutes for preparing bread without gluten (*5*). When xanthan gum was used as a network in the preparation of cornstarch bread, the resulting product had good specific volume but a coarse crumb texture and lack of flavor (*6*). The use of hydroxypropylmethylcellulose (HPMC) as a substitute for gluten in a rice bread formula seems to be the best alternative to provide the gas-retaining and structure-forming properties in the crumb (*7*, *8*). In fact, a comparative study using different gums (HPMC, locust bean gum, guar gum, carrageenan, xanthan gum, and agar) in a rice bread formulation showed that HPMC resulted in the highest specific loaf volume (*9*). A different approach for improving the quality of the gluten-free breads has been to blend the rice flour with different flours such as soy flour and starches such as corn and cassava starches in order to obtain good loaf volume and a uniform crumb without large holes (*10*).

Those previous studies have allowed rice flour breads with loaf specific volume comparable to that of wheat breads to be obtained, although some improvement in sensory appearance and crumb texture is still required. This further improvement becomes very complicated due to the hydrophobic nature of the rice proteins compared to the hydrophilic nature of the wheat gluten (*11*). A quality increase of wheat bread is reached by

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the addition of different enzymes (amylases, carbohydrases, phytase, and lipase) and also hydrocolloids (*12*−*14*); however, rice flour is less responsive to the presence of dough conditioners and enzymes (*7*). Therefore, new conditioners and enzymes are needed as rice bread improvers.

The objectives of the present investigation were to explore the effects of cyclodextrin glycosyl transferase or cyclodextrinase (CGTase) (EC 2.4.1.19) in dough rheology and bread quality obtained from rice flour. CGTase is able to catalyze four different reactions: i, cyclization; ii, coupling; iii, disproportionation; and iv, hydrolysis (*15*). Some of the most interesting products of those reactions are the cyclodextrins, which are made from the hydrolysis and cyclization of starch obtaining closed circular molecules of six, seven, or eight glucose units and referred to as α-, β-, or γ-cyclodextrin, respectively. Those molecules have a polar surface responsible for the aqueous solubility and a hydrophobic inner core. Due to the special properties of the cyclodextrins, CGTase is currently used for the production of different foods and other materials (*16*, *17*). In the case of rice bread, due to the hydrophobic nature of the rice proteins, the use of CGTase could reduce the hydrophobic environment by hydrolyzing and cyclizing the starch and also through the hydrolysis products that can form complexes with a variety of solid, liquid, and gaseous compounds.

MATERIALS AND METHODS

Materials. Commercial rice flour from Huici Leidan S.A. (Navarra, Spain) was used in this study. The particle size of the rice flour was compressed to between 177 and 37 μm. The rice flour had moisture, ash, and protein contents of 12.8, 0.57, and 8.83%, respectively, and the amylose content was 21.9%. HPMC Methocel K4M from Dow Chemical Co. (Midland, MI) was used. CGTase (3.3 units/mL) and α-amylase (Novamyl, 1500MG) were kindly provided by Novo Nordisk (Madrid, Spain). α-Cyclodextrin, β-cyclodextrin, and γ-cyclodrextrin standards were obtained from Sigma-Aldrich (Sigma-Aldrich, St. Louis, MO). Vegetable seed oil, compressed yeast, sugar, and salt were obtained from the local market. All reagents were of analytical grade.

Bread-Making. Initial trials were carried out to produce acceptable rice bread according to the formula given by Nishita et al. (*7*) at water absorption of 75%. At this water absorption the dough consistency was very high (500 Brabender units, BU) and the bread dough showed very little rise during proofing at 30 °C and 80% relative humidity; as a consequence the resulting bread had very poor specific volume (1.10 mL/g). Increasing the water content to >75% resulted in better proofing and loaf breads of higher specific volume. Water absorption of 90% was found to be optimum and was selected for the subsequent study. Rice flour (500 g) and HPMC (variable) were blended and added to the bowl of the Hobart mixer (N50, Hobart, Canada) containing sugar (7.5%, flour basis), salt (2%, flour basis), yeast (3%, flour basis), and CGTase (variable) dissolved individually in parts of 450 mL of water. Oil (variable) was added, and all of the ingredients were mixed for 1 min at speed 2 with the paddle attachment followed by mixing for 30 s at speed 3. The dough was scraped down and again mixed for 2 min at speed 2, scraped, and mixed for 30 s at speed 3. A constant weight of dough (100g) was scaled in well-greased pans (measuring 70 × 40 mm), proofed for 60 min at 30 °C and 80% relative humidity, and then baked at 175 °C for 40 min. Bread was removed from the pan and cooled on a rack for 1 h.

When the effect of α-amylase was tested, the enzyme (0.02%, w/w flour basis) was added to the rice bread recipe. This enzyme dosage was the one used in bread-making (*18*).

The farinograph (Brabender, Duisburg, Germany) was used to determine the dough consistency. Mixed dough (106.2 g) was placed in the 50 g bowl of the farinograph, and the dough consistency was reported after 5 min of mixing.

Bread Quality Evaluation. The following characteristics were assessed: weight, volume (rapeseed displacement), specific volume, and width/height ratio of the central slice (shape index). Crumb firmness was determined on the Texture Analyzer TA-XT2i (Stable Micro Systems, Surrey, U.K.) after 24 h of baking. A bread slice of 20 mm thickness was compressed to 50% of its original height at a crosshead speed of 1 mm/s with a cylindrical stainless steel probe having a diameter of 25 mm. The peak force of compression was reported as firmness (g). The crumb samples were also freeze-dried and stored in airtight plastic bottles for further analysis for cyclodextrins.

Cyclodextrin Analysis. The amount of cyclodextrins released as a consequence of the CGTase activity was determined in the freeze-dried samples. Samples (100 mg) were suspended in 1 mL of distilled water, vortexed for 10 min, and then centrifuged at 15700*g* for 5 min. The clear supernatant was filtered through 0.22 μm membrane filters and used for HPLC analysis. The liquid chromatography was performed in a Hewlett-Packard HP1050 including the following components: high-pressure reciprocating pump, injection valve, Rheodyne model 7125, with a 5 mL injection loop; chromatographic column, Tracer NH_2 (25 × 0.4 cm, Spherisorb, 5 μm); column oven, Waters, model TCM; refractive index detector, Hewlett-Packard model 1047A; and integrator/plotter, Shimadzu model Chromatopac C-R3A. Samples were eluted with acetonitrile/water (65:35, v/v) at 1 mL/min. The column and flow cell were set at 40 °C. The cyclodextrins (CD) (α, β, and γ) were used as reference standards.

Viscosity and Rheological Analysis. The Rapid Visco analyzer (Newport Scientific Pvt. Ltd., Warriewood, Australia) was used to determine the pasting properties of the rice flour and to study the effect of CGTase on those properties. Pasting properties were determined following the standard Newport Scientific rice method. Rice samples were run in duplicate.

Dynamic rheological measurements were performed on a controlled stress rheometer (Rheostress 1, Thermo Haake, Karlsruhe, Germany). The rice dough was prepared by mixing rice flour (50 g) along with enzyme and 45 mL of water in the farinograph. The mixing was carried out for 15 min after the addition of water, and then the dough was immediately sealed in an airtight plastic container. The rice dough was placed between parallel plates (60 mm diameter), and the gap was adjusted to 1 mm. Vaseline oil was used to coat the outer edges to prevent drying of the sample. The dough was allowed to rest for 5 min so that residual stresses could relax. A frequency sweep from 0.01 to 10 Hz was performed at a constant stress of 2 Pa at 30 °C. Preliminary trials were carried out to determine the stress at which the sample remained in the linear viscoelastic region. This was important because the evaluation of the physical properties in the linear range is easier and more reliable and the low stress applied is not injurious to the dough structure. The dough structure was evaluated by comparing log log plots of G' and G'' with frequency.

Statistical Analysis. A central compound rotational type design with 18 treatments (8 factorial points, 6 axial, and 4 repetitions of the central point) was used. The three variables had values of X_1 (CGTase, 0, 10, 20, 30, and 40 μL/100 g of flour), X_2 (HPMC, 2, 3, 4, 5, and 6 g/100 g of flour), and X_3 (oil, 2, 4, 6, 8, and 10 mL/100 g of flour). The following equation was used:

$$Y = a + \sum_{i=1}^{3} b_i X_i + \sum_{i=2}^{3} c_i X_1 X_i + \mathrm{d} X_2 X_3 + \mathrm{e}\, X_1 X_2 X_3 + \sum_{i=1}^{3} f_i X_i^2$$

RESULTS AND DISCUSSION

Dough Consistency and Dynamic Rheology. Dough consistency increased with increasing HPMC concentration (**Table 1**) and was most significantly affected by HPMC concentration (**Table 2**). The consistency increased by 120% with increase in HPMC from 2 to 6%. Conversely, oil lowered the consistency by 20% with increase in levels from 2 to 10%. The increase in consistency with increasing levels of HPMC can be attributed to the ability of HPMC to bind large amounts of water (*19*), whereas oil acts as a lubricant between the particles in the dough and decreases resistance to mixing, lowering dough consistency. The addition of CGTase lowered dough consistency with increase in level from 0 to 40 μL, suggesting that it was bringing

Table 1. Experimental Design and Responses

sample	CGTase (μL/100 g of flour)	HPMC (%, fb)[a]	oil (%, fb)[a]	dough consistency (BU)[b]	specific volume (cm^3/g)	shape index	crumb firmness (g)
1	10	3	4	280	2.1	1.078	587.2
2	30	3	4	280	2.0	1.175	305.9
3	10	5	4	365	2.1	1.018	720.8
4	30	5	4	380	1.8	1.063	655.2
5	10	3	8	245	3.4	0.842	240.4
6	30	3	8	240	3.2	0.865	190.5
7	10	5	8	370	4.6	0.732	175.0
8	30	5	8	370	3.8	0.733	173.4
9	0	4	6	330	2.5	0.805	314.0
10	40	4	6	280	3.8	0.777	209.0
11	20	2	6	200	2.7	0.923	308.6
12	20	6	6	440	4.6	0.703	399.3
13	20	4	2	350	1.5	1.086	388.3
14	20	4	10	280	4.3	0.725	201.0
15	20	4	6	325	4.2	0.775	263.7
16	20	4	6	320	4.5	0.774	248.0
17	20	4	6	320	4.2	0.773	231.1
18	20	4	6	330	4.3	0.774	247.1

[a] fb, flour basis. [b] BU, Brabender units.

Table 2. Regression Coefficients[a]

interaction	farinograph consistency	specific volume	shape index	crumb firmness
constant	199.50	−4.072	1.623	1412.506
X_1	0.017	0.024	0.002	−11.402*
X_2	7.35*	0.185	−0.022	−40.839
X_3	−3.275	0.164*	−0.024	−18.192
X_1^2	−0.002**	−0.00012**	4.783×10^{-6}	0.003
X_2^2	−0.09*	−0.0077*	0.0007*	1.304*
X_3^2	−0.035*	−0.0037**	0.0004*	0.177
X_1X_2	0.025	0.0003	−0.0001	0.383
X_1X_3	0.005	0.0003	-7.237×10^{-5}	0.241
X_2X_3	0.225*	0.0075*	−0.0004	−0.575
$X_1X_2X_3$	−0.0005	-2.3×10^{-5}	2.025×10^{-6}	−0.008
R^2	0.974	0.834	0.808	0.863

[a] X_1, X_2, and X_3 are CGTase, HPMC, and oil, respectively. *, **, significance at $p < 0.5$ and $p < 0.05$, respectively.

about some breakdown in the starch during the mixing process. All three variables significantly affected the consistency in square terms, with CGTase having the most significant effect ($p < 0.05$). The interactive effects of HPMC and oil were also significant ($p < 0.5$). The regression model has an R^2 of 0.974 and can be used to predict the dough consistency when the three ingredients are present at different levels.

Dynamic rheological studies of the rice flour dough showed that the elastic modulus was higher than the viscous modulus, which suggests a solid elastic-like behavior of the rice flour dough. Both the elastic (G') and viscous (G'') moduli increased with increasing HPMC concentration (**Figure 1**). The magnitude of the modulus increased with increasing HPMC, indicating that the dough became stronger as HPMC increased (results not shown). The elastic modulus remained higher than the viscous modulus at all of the concentrations studied. The difference between elastic and viscous modulus became less and the modulus became more frequency-dependent as HPMC concentration increased, indicating increasing viscous-like behavior (results not shown). Addition of oil increased both the elastic and viscous moduli, but the increase in the elastic modulus was higher, suggesting that oil contributed more to the elastic behavior of the dough. Incorporation of CGTase to the dough lowered the elastic modulus and complex viscosity of the dough but did not seem to affect the viscous modulus. The

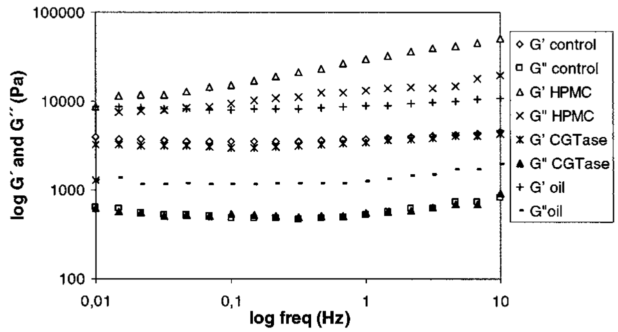

Figure 1. Dynamic rheological data of rice flour dough as affected by CGTase (40 μL/100 g flour), HPMC (6%, flour basis), and oil (10%, flour basis).

tan δ (G''/G') increased in the presence of the enzyme, suggesting that the relative contribution of the solid character (G') decreased. Although the optimum activity of the CGTase is reported to be ∼70 °C, it must be acting on the damaged starch during the mixing process and also during the proofing period (30 °C), bringing about some hydrolysis, which affects the dough rheology. A decrease in the elastic modulus and an increase in the tan δ in wheat flour dough from sprouted wheat flours have been reported by Singh et al. (*20*) and were attributed to higher amylase and protease activities.

Effect of CGTase on Rice Bread Quality. Preliminary trials revealed that CGTase had positive effects on rice bread volume and that varying the HPMC and oil levels in the formula affected the bread specific volume. Therefore, it was decided to study the combined effect of CGTase, HPMC, and oil on rice bread quality by following an experimental design (**Table 1**).

Increasing levels of all three ingredients increased the specific volume (**Table 1**). The specific volume positively correlated with the bread volume with a correlation coefficient of 0.99. Oil concentration significantly affected specific volume (**Table 2**), which increased by almost 3 times with an increase in oil from 2 to 10%. Increasing levels of fat have been reported to improve bread volume in wheat flour bread (*21*). Increasing HPMC concentration from 2 to 6% increased specific volume by 67.64%. This is attributed to the HPMC ability of retaining CO_2 by decreasing the permeability of the dough to CO_2 (*12*). The interactive effect of HPMC and oil also significantly affected specific volume ($p < 0.5$). Increasing the CGTase concentration from 0 to 20 μL/100 g of flour increased the specific volume by 73%. Likely this effect is due to the release of fermentable sugars as a consequence of the hydrolysis activity of the CGTase, because this enzyme has the ability to hydrolyze starch as the α-amylase does. These results agree with the rheology data previously presented, where even at 30 °C the presence of CGTase produced a decrease in the elastic modulus of dough attributed to α-amylase and protease activities (*20*).

Because α-amylase is the most widely used enzyme in the baking of wheat bread due to its effect on bread volume and crumb texture, a comparison study was carried out to establish the potential use of CGTase as a gluten-free bread improver. When α-amylase was added to the rice bread recipe using the optimum amounts of HPMC (4%) and oil (6%), the resulting bread had a specific volume of 3.3 mL/g, which was higher than the one obtained in the absence of CGTase (2.5 mL/g) but lower than the specific volume obtained with the addition of 20 μL/100 g of flour of CGTase (4.3 mL/g). Therefore, the effect of CGTase on the specific volume of rice bread cannot be completely explained in terms of production of fermentable sugars.

Table 3. Influence of the CGTase and Oil (6%) on the Pasting Properties of Rice Flour Determined with the Viscoanalyzer[a]

sample	peak (cP)	trough (cP)	breakdown (cP)	final viscosity (cP)	setback (cP)
control	2783 ± 11	1625 ± 16	1159 ± 18	3295 ± 21	513 ± 17
CGTase (20 μL/100 g of flour)	2424 ± 21	1174 ± 10	1250 ± 24	3147 ± 14	723 ± 23
CGTase (40 μL/100 g of flour)	2136 ± 13	737 ± 15	1399 ± 11	2027 ± 6	−109 ± 16
oil	2844 ± 11	1910 ± 21	934 ± 16	3420 ± 18	576 ± 13
CGTase (20 μL/100 g of flour) + oil	2458 ± 18	1471 ± 16	987 ± 8	2620 ± 22	162 ± 11

[a] Means ± standard deviation ($n = 3$).

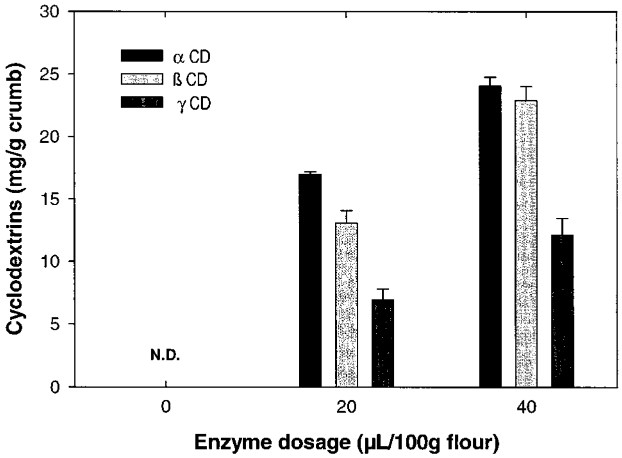

Figure 2. Composition of cyclodextrins (CD) isolated from loaves treated with different amounts of CGTase. ND indicates that no cyclodextrin was detected.

All three variables (CGTase, HPMC, and oil) lowered the shape index (width/height), which describes the loaf shape. A shape index value of close to 1 indicated that the height and width of the bread loaf were same and it has a low volume, whereas shape index values of close to 0.7 indicated loaves of higher volume. Therefore, the three variables improved the loaf shape.

With regard to the crumb texture, all three variables lowered crumb firmness with effects of CGTase being most significant in both linear and square terms (**Tables 1** and **2**). The crumb firmness showed a negative correlation with specific volume. The addition of increasing CGTase concentration considerably lowered firmness by 53%, obtaining a very soft bread crumb. The same reduction in the crumb firmness was obtained with the addition of α-amylase, but crumbs were very sticky (data not shown). These results indicate that the improvement produced by the addition of CGTase might be due to the starch hydrolysis that yields fermentable sugar but also to the cyclization of the hydrolysis products, which could form complexes with lipids and also proteins.

Cyclodextrins Produced by CGTase. CGTase has multiple catalyzing activities; therefore, the rice bread improvement could result from the combined effect of those activities.

Although CGTase has a cyclizing activity, it is necessary to confirm that the bread-making conditions are favorable for that reaction to take place. The amounts of the different cyclodextrins produced during bread-making are presented in **Figure 2**. No detectable amount of cyclodextrins was observed in the crumb from rice bread obtained in absence of CGTase. The cyclodextrin concentration increased by increasing the CGTase dosage. α-Cyclodextrin was produced to greater extent, followed by *β*- and *γ*-cyclodextrins, although at the highest enzyme concentration tested (40 μL/100 g of flour) the *β*-cyclodextrins underwent greater increase compared with the other cyclodextrins, because CGTase produces predominantly α-cyclodextrins in the earlier stage of reaction and at low enzyme concentration; however, with prolonged reaction time or high enzyme dosage the amount of *β*-cyclodextrins exceeds that of α-cyclodextrin (*22*, *23*). The presence of cyclodextrins confirms the cyclizing activity of CGTase during the bread-making process.

Therefore, during bread-making the hydrolyzing activity of the CGTase produces the same effect as the α-amylase, that is, the starch hydrolysis yielding fermentable sugars, which are metabolized by yeast, and consequently high loaf volume and soft crumb are obtained. Second, the reaction of cyclization yields cyclodextrins, which have the ability to form complexes with solid, liquid, and gaseous compounds. In fact, cyclodextrin molecules form inclusion complexes with triglyceride molecules and lipids, lowering interfacial tension, and thus act as emulsifiers (*24*, *25*); the softening effect of the emulsifiers on the crumb of wheat bread is well-known (*26*). In addition, cyclodextrins have the ability to interact with hydrophobic proteins, leading to increased solubility (*27*). The cyclodextrins may be forming complexes with the hydrophobic proteins (globulin and glutelin) of rice, increasing their solubility. These complexes may be involved in film forming and better entrapment of the CO_2, leading to increased volume and better texture.

Pasting Properties of Rice Flour As Affected by CGTase. Because the pasting properties of rice flour have been reported to be very useful in predicting the bread-making properties (*28*), it was decided to analyze the influence of CGTase addition on the gelatinization behavior of rice flour in order to explain the effects of the CGTase on bread texture. In **Table 3**, it can be seen that the addition of CGTase (20 μL/100 g of rice flour) lowered the peak viscosity (2424 cP) and slightly affected the final viscosity (3147 cP), indicating that the enzyme acts on the starch, breaking it and thus lowering the viscosity. CGTase also increased the breakdown (1250 cP), indicating that the paste was less resistant to heating and shear stress because starch had been hydrolyzed. When added at higher levels (40 μL/100 g of rice flour), a further decrease of the peak viscosity (2136 cP) along with an increase of the breakdown (1399 cP) was observed. The setback is the difference between the peak viscosity and the viscosity at 50 °C. This parameter is related to the retrogradation behavior of starch, which in turn is related to amylose helix interaction, and is one of the most important parameters in predicting rice bread characteristics (*28*). In fact, soft bread crumbs are obtained from rice flour with low viscosities at 50 °C and low setback values, whereas high viscosities at 50 °C and high setback values give harsh texture (*28*). The addition of CGTase decreased the final viscosity, although the setback was reduced only when the highest enzyme dosage tested (40 μL/100 g of flour) was added. Probably at low enzyme dosage some hydrolysis products could also associate during cooling as the amylose does, and at high enzyme concentration a high amount of cyclodextrins is present, which could physically interfere with the amylose complex as has been suggested by Liang et al. (*25*). From the effect of the highest concentration of CGTase on the gelatinization behavior of rice flour, it could be predicted that softer crumb texture might be obtained by adding this enzyme, which agrees with bread-making results.

The cyclizing activity of the CGTase might promote an additional effect on the gelatinization behavior of rice flour, because the resulting cyclodextrins could form complexes with different compounds, for instance, the lipids added in the rice bread recipe. This was confirmed by results shown in **Table 3** for the effect of CGTase combined with oil. The addition of oil to rice flour slightly increased the peak and final viscosities and also the setback, whereas a considerable decrease was observed in the breakdown, likely due to the inhibition of the amylose leaching (*29*). When a combination of CGTase and oil was added to rice flour, the effect during heating was the sum of the individual effects, but a great synergistic effect was observed during the cooling cycle. The addition of CGTase in the presence of oil produced a pronounced decrease in the final viscosity and setback, which indicates the interaction between the lipids with the cyclodextrins and other hydrolysis products obtained from the enzyme reaction. This result agrees with previous findings of Shimada et al. (*24*) and Liang et al. (*25*), who determined the inclusion complexes formed among cyclodextrins, triglyceride molecules, and lipids.

Conclusion. The improvement of the rice bread by the traditional bread-making conditioners and improvers is very difficult due to the hydrophobic nature of the rice proteins. The enzyme cyclodextrin glycosyl transferase, due to its multiple catalyzing activities, seems to be a good improver for rice bread quality. Rice bread with good specific volume and very soft crumb texture is obtained by the addition of CGTase. Likely the products from the enzyme reaction can form complexes with the rice proteins, decreasing their hydrophobic nature, and also with lipids and other compounds present in the bread-making process. By using this enzyme it is possible to obtain rice bread that actually meets the quality expectations of the gluten-free consumers.

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