

Influence of storage time and starches from *Pueraria lobata* (Kudzu) root and *Zea mays* (Corn) on aroma, basic taste, and flavor attributes of conventional milk yogurt

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Article history

Received: 1 July 2014

Received in revised form:

3 December 2014

Accepted: 12 December 2014

Abstract

This research compared the effects of two types of starch (corn and kudzu) at different concentrations on the sensory attributes (aroma, basic taste and flavor) of conventional milk yogurt. The effects of days of storage, starch type and concentration, and their interactions on 40 sensory variables were determined by the use of covariate analysis. Time and starch interaction were significant for only 1 sensory response starch. 34 sensory responses had a time-dependent response, while 6 variables were affected by starch. The sensory variables affected by time were: 15 of 17 aromas, 6 of 6 basic tastes, and 13 of 17 flavor attributes. Best-fit polynomial regressions over time were determined for the responses affected. Response surface methodology (RSM) was applied to determine the effects of starch type and concentration on yogurt products. Plain yogurt with added kudzu or corn starch has a potential to succeed in the market. The use of kudzu starch may provide a smooth and bright appearance and good flavor in fortified yogurt.

Keywords

Yogurt

Kudzu starch

Corn starch

Sensory

Aroma

RSM

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Introduction

Kudzu *Pueraria lobata* (*Pueraria montana* var. *lobata*, *Pueraria thunbergiana*, *Pueraria lobata* Willd.) is one of the species of *Pueraria* in the pea family *Fabaceae*. It is a plant from southern Japan and southeast China. Two of its isoflavones, daidzin and daidzein, are the major antidipsotropic (anti-alcohol abuse agent) constituents of kudzu (Overstreet *et al.*, 1996; Keung, 1998; Keung and Vallee, 1998; Rong, 1998). *P. lobata* root was also used for the treatment of hypertension (Keung, 2002) and alcoholism (Lin *et al.*, 1996), and as an antioxidant (Guerra *et al.*, 2000). Kudzu root starch is a traditional natural starch widely used in Japan for its superior thickening properties.

Corn (*Zea mays*) provides a high-quality, inexpensive starch used widely in the food industry for the development of various textures and viscosity (White and Yao, 2007). Clarity, flavor, and durability to withstand rigorous processing conditions are also common requirements (Whistler *et al.*, 2009). According to Geng *et al.* (2006), kudzu starch and corn starch have similar amylose contents ($P > 0.05$). Kudzu and corn starch had similar thermal properties. Spherical, hemispherical, and polygonal-shaped

granules can be seen for kudzu starch. The corn starch granules are known to have pores or channels on the surface with a polygonal shape (Geng *et al.*, 2006). Starch is used in yogurt to increase its viscosity, improve its mouth-feel, and prevent syneresis (Doreau, 1995; Hess *et al.*, 1997; Williams *et al.*, 2003; Williams *et al.*, 2003; Tamime and Robinson, 2007; Ares, 2007). Starch is one of the most widely used thickening agents in yogurt production due to its processing ease and low cost when compared with other hydrocolloids (Foss, 2000).

Yogurt consumption has changed over the past several decades in the United States. Acceptance of plain yogurt depends on acidity, aroma, flavor, and texture. Long-term storage of yogurt causes an off-flavor because of the production of undesired aldehydes and fatty acids. Since there is an association between the sensory properties of yogurt and the volatile flavor compounds, more research studies are needed to clarify the relationship between yogurt aroma and flavor with volatile compounds (Cheng, 2010). Several factors account for the success of yogurt, including its natural appearance and its organoleptic characteristics (fresh, acidulated taste and characteristic flavor) (Akin and Akin, 2005).

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Taste, flavor, consistency, and color of the skimmed and whole milk yogurts showed acceptable values over the storage time (Salvador and Fiszman, 2004). Plain probiotic yogurt made from skimmed and whole milk powder has been stored in a refrigerator at 4°C for 35 days. They also found no significant changes in color, flavor intensity, or sweetness for both yogurt types (skim and whole milk yogurt) after a storage period (Obi *et al.*, 2010). Different sensory aroma descriptors have been used to characterize the aroma of yogurt (Coggins *et al.*, 2008; 2010). Kudzu could be an alternative starch to use in yogurt to improve the quality of the sensory properties of yogurt, such as textural properties, flavor, aroma, and basic taste. Due to the isoflavonoids content and antioxidant activity of *Pueraria lobata*, it can be applied to yogurt as a functional food in the industry.

To our knowledge, kudzu starch has not been studied in yogurt. The purposes of this study were to investigate a sensory analysis of *Pueraria lobata* (kudzu) root starch in conventional milk yogurt, and to determine the sensory properties of yogurt with kudzu root starch and corn starch.

Materials and Methods

The commercial Japanese kudzu root starch was obtained from Wild Organic Kudzu (Mitoku Co., NC), and the corn starch (stabilizer; Dairyblend TG-AG) was obtained from TIC Gum Company.

Yogurt milk preparation

The yogurt base was made with skim milk, cream, nonfat dry milk (low-heat Grade A nonfat milk; Danish Creamery Assoc., Fresno, CA), and stabilizer (Continental Colloids, Inc., West Chicago, IL). The yogurt milk was formulated by adjusting skim milk to 1% milk-fat using 36% cream. The skim milk and cream were obtained from the Custer Dairy Processing Plant at Mississippi State University. Nonfat dry milk (NDM) (Danish Creamery Assoc., Fresno, CA) was added at a rate of 4% along with 1%, 2%, 2.5%, or 3% w/w (Bourgouin, 1993) commercial stabilizer (modified food starch, carrageenan, and pectin; Continental Colloids, Inc., West Chicago, IL, US). The same procedure was repeated using kudzu root starch (Mitoku Co., NC) at the same rates. These ingredients were mixed, heated (at 50°C), and homogenized using a HTST system (Model #-AT20 DF, Paul Mueller Company, Springfield, MO) (Tamime and Robinson, 2007). The processing of yogurt milk was repeated to produce three batches.

Yogurt production

Yogurt was stored for 49 days for the study. The yogurt production procedures of Coggins *et al.* (2010) were utilized. The yogurt milk batch was heat treated at 85°C for 30 min in a 189-L stainless steel vat (Walker) (Tamime and Deeth, 1980; Tamime and Robinson, 2007), followed by rapid cooling to 42°C in an ice water bath. The warm yogurt milk was then inoculated with a yogurt culture (YC-087; Chr. Hansen Laboratory, Inc., Milwaukee, WI) that contained strains of *Streptococcus thermophilus* and *L. delbrueckii* ssp. *bulgaricus*. The incubation was terminated when the pH decreased to levels between 4.2 and 4.5; then, it was cooled to 10°C and stirred in the vat. During the incubation, the temperature of the fermenting milk did not drop below 39°C, which is an industry standard. Using the same procedure, a control yogurt was made every 7 days, up to day 49, for the descriptive studies.

Yogurt treatments/experimental design

Yogurt samples were produced for the two starch types at four concentrations in three batches (replications). All formulated samples were immediately refrigerated at 5°C and stored from 0 to 49 days. A control yogurt without corn or kudzu starch was also produced.

Storage of yogurt

The stirred yogurt samples were scooped into 170 ml polypropylene cups (Landis Plastics, Inc. Chicago Ridge, IL), and the lids were sealed. The samples were immediately stored in well-ventilated incubators at 5°C (BOD30A14, GS LabE9, Revco, Asheville, NC). Prior to sensory evaluation, the yogurt samples were removed from the incubators, placed on plastic trays, and taken to a sensory preparation area within 10 min. The trays had a 340 ml opaque plastic cup (Sysco, Memphis, TN) of distilled water, a 340 ml expectorate cup (Sysco, Memphis, TN), unsalted crackers (Premium Brand, Nabisco), a sensory instrument, white plastic spoons (Sysco, Memphis, TN), and napkins (Kroger Brand, Cincinnati, OH). Trays with the yogurt samples were randomly assigned to the panelists for evaluation.

Sensory evaluation of yogurt

Descriptive terms were generated for this study by the panelists from individual evaluations of the yogurt. Samples were evaluated for aroma, basic taste, and flavor. The procedures of Coggins *et al.*, (2008, 2010) were utilized in this study.

The yogurt samples stored at 5°C were evaluated on days 1, 7, 14, 21, 28, 35, 42, and 49. For scoring,

a 15-point intensity scale (where 0 = not detected and 15 = extremely strong) was used. For each session, 12 samples (4 different concentrations in 3 replicates or batches) were tested per session, and the sessions were scheduled with three sensory evaluations per month. The panel was trained in the use of the chosen attributes (Coggins *et al.*, 2008) for 72 h over a 6-month time period (3 h/week). The yogurt cups, brought to ambient temperature, were served with water (Magnolia Springs, MS) and unsalted crackers (Premium brand; Nabisco Foods; Subsidiary of Kraft Foods, Glenview, IL), which were used as palate cleansers between samples. The testing was performed in a sensory laboratory equipped with individual booths and artificial daylight (fluorescent). Panelists were given freshly made (day 1) plain yogurt samples without any starch prior to sensory evaluation. Samples were run in triplicate with a randomized order of presentation to the panelists. The yogurt samples were identified by a three-digit random number code. Expectation of each sample was encouraged. Formal descriptive round table forums were completed on the yogurts. Approval for this research was from IRB Docket # 09-126. Protocol was followed for Human Subject Research (Mississippi State University Internal Review Board procedures specifying work with human subjects).

Statistical analyses

Statistical analysis was executed using the procedures of SAS 9.1 (SAS, 2003). Covariance analysis (PROC MIXED) was executed on each sensory attribute to test for a significance of the effects of two starches, at four concentrations, evaluated for 1 to 49 days and their interactions. When the interactions of two variables were significant, data was further analyzed by holding one variable constant while analyzing for the response of a second variable. If attribute values changed with time, response surface methodology (PROC RSREG) was performed to simultaneously characterize the response of all concentrations of kudzu or corn over time. Response surface methodology can be difficult and if the results were not satisfactory for any attribute, that attribute was then analyzed as a polynomial regression over time for each level of kudzu concentration (PROC REG). With numerous statistical tests, the significance level is set at P= 0.01 to control Type I error.

Polynomials fit to attribute values were linear, quadratic, or cubic. The best model was determined as a function of non-reducing adjusted R-square value and the parsimony principal. Curves, such as a linear time response for 1% kudzu starch and

a linear time response for 1% corn starch, were compared where appropriate, in order to see if they were significantly different by using the general linear model's significance of residual sum of squares approach (Ryan, 2007).

Results and Discussion

Aroma

An analysis of variance for the significance of starch type, concentration of starch (linear and quadratic), days of storage (linear and quadratic), and their interactions shows that 2 of the 17 aroma attributes, yeast and caproic acid, were unaffected by any source of variation. All other attributes had a significant storage time response; no attribute was affected by the type of starch; and two attributes, potato and overall intensity, were significantly affected by the concentration of starch (Table 1).

Table 1. Regression equations describing the effects of days of storage (day) and concentration of starch (concentration) on aroma attributes (Regression equation: a+bDay+cDay²+dConcentration)

ATTRIBUTE	Constant number	Day	Day ²	Concentration
Overall intensity	6.62	-0.074	+0.0001	-0.168*
Sour	4.65	-0.077	+0.0015	
Sour cream	3.94	-0.153	+0.0028	
Cream	2.41	-0.031		
Powdered milk	1.56	-0.062	+0.001	
Milk (whole)	1.95	-0.03		
Band aid®	1.41	-0.088	+0.002	
Butter milk	2.86	-0.112	+0.002	
Rancid	0.12	-0.011	+0.0004	
Plastic	0.35	-0.025	+0.0006	
Valeric acid	0.44	-0.026	+0.0006	
Potato	0.83	-0.053	+0.0008	+0.077
Whey	2.5	-0.026		
Cheesy	1.14	-0.016		
Baby vomit	0.734	-0.04	+0.0013	

*All regressions have a P≤.01 level of significance

Yeast and caproic acid are not typical fresh yogurt aromas, and they were not perceived to change during storage. The intensity of potato aroma is a result of volatile loss, ingredient interactions, syneresis, and other factors affected by concentration. Since attributes such as overall intensity and potato were affected by two quantitative variables simultaneously, the attribute intensities were plotted as a response

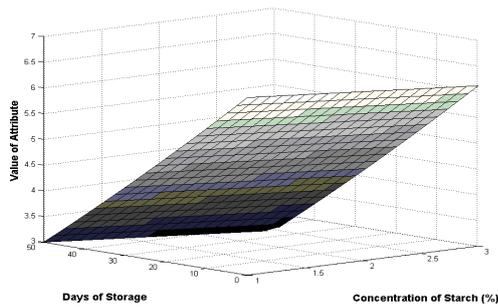


Figure 1. Response surface for overall intensity of aroma attribute for yogurt with kudzu/corn starch with respect to day and starch concentration (0-3%)

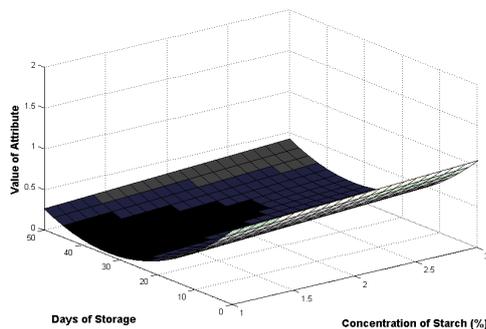


Figure 2. Response surface for potato of aroma attribute for yogurt with kudzu/corn starch with respect to day and starch concentration (0-3%)

surface (Figure 1 and 2). For overall intensity, Figure 1 shows the greatest on the first day at the lowest starch concentration. Figure 2 is the response surface for potato aroma scores affected by quadratic days of storage response and a linear effect of concentration of starch concentration. In Figure 2, the maximum value of potato aroma attribute was near 1.25 at day 0. The attribute value dropped to near 0 over days and increased slightly near day 49, but concentration early all values of this attribute were near zero. The response increases slightly for an increasing concentration of starch, but it decreases rapidly over time with a very slight quadratic curvature. The intensity of potato attribute (Figure 2) is greatest at the beginning of the study for the highest starch concentration. This intensity decreases to a minimum near 30 days of storage, and then it increases (Figure 2). For potato, the intensity is between 0 and 1 and varies little in absolute terms. The 13 attributes that are only affected by days are of two types (Figure 3). First are the linear responses, which decrease over time, and second are the quadratic responses, which have a minimum on days 14 to 28 and then begin to increase. Some responses – such as plastic, rancid, valeric, powdered milk, and cream – have intensity values that varied little from near zero. Even though

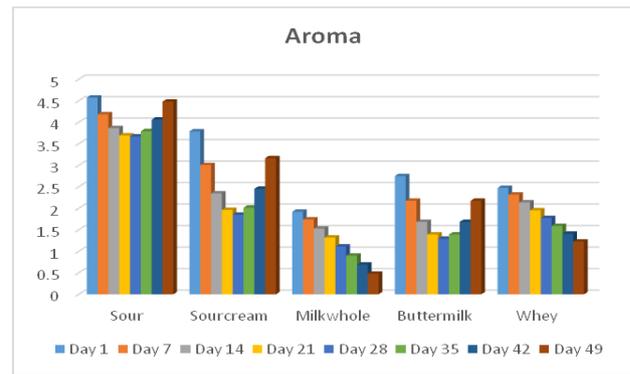


Figure 3. Effect of time on aroma attributes of conventional yogurt with starches (kudzu/corn)

some of these attributes are related and contain identical volatiles in their compositions, the panelists discerned little change in these values over days of storage.

There are several factors, such as storage, temperature, microbial factors, source of milk, added flavors or thickeners, and other factors affecting the overall yogurt aroma (Routray and Mishra, 2011; Prasad, 2013). The presence of starch in yogurt was found to induce a significant decrease of aroma compounds in the headspace, as observed by Decourcelle *et al.* (2004). Because of the difference of interaction of various types of thickeners with the yogurt matrix, kudzu or corn starch may have affected yogurt aroma in this study. Kora and others (2004) found that the thickening agent and mechanical treatment had little influence on aroma compound retention.

Basic taste

The analysis of variance testing for the significance of three sources of variation – type of starch, concentration of starch (linear and quadratic), and days of storage (linear and quadratic) – and their interactions showed that the days of storage were highly significant for all basic attributes except for salty, which was unaffected by any source of variation (Data Table is not showed). Only one of the six attributes, sour, was significantly affected by the type of starch. With aroma attributes, none of the interactions among main effects were significant. All attributes have a quadratic response to days of storage except for sour, which has a common linear response with different intercepts for each type of starch. The intensity of sweet was near zero over days of storage (Data Figure is not showed). Values for the attributes prickly and bitter were only slightly greater. The astringency increased in intensity for increasing days of storage with a maximum at about day 40. It was reported that storage was found to have a significant effect on pH, acidity, syneresis, basic taste, and

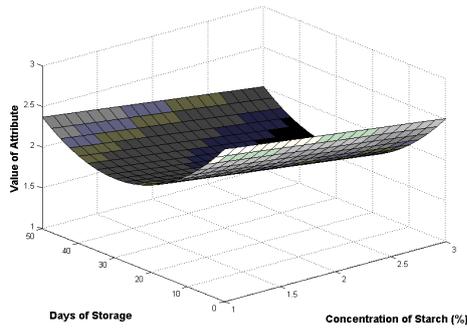


Figure 4. Response surface for whey of flavor attribute for yogurt with corn starch with respect to day and starch concentration

texture ($P < 0.05$) (Routray and Mishra, 2011; Prasad, 2013).

Flavor

The maximum score value of rancid flavor attribute was 0.6 at day 42 and at highest concentration 3 (3% kudzu and corn starch). The response surface for attribute rancid (Data Figure is not showed) shows the value near zero on day 1 but increasing after about day 20 to values between .5 and .75 by day 49. Rancidity is an attribute depending on extended storage, handling, temperature abuse or old ingredients into mix. The study was carefully monitored and the factors were associated with none of the above with the exception of extended storage. It was demonstrated response surface plot for rancid of flavor attribute for yogurt with kudzu/corn starch.

As shown in Figure 4, a concentration x day interaction had an effect on predicted whey attribute with corn starch. The predicted value was maximum value (2.9) at day 1 and 1% starch and decreased until day 28 days and increased slightly until day 49 depending on concentration of corn starch concentration. Whereas corn starch in yogurt exhibited day and concentration effect on whey flavor attribute (Figure 4), kudzu starch-yogurt product showed a linear day effect.

The analysis of variance testing for statistical significance of starch type, concentration of starch, days of storage, and their interactions on flavor attributes showed a complexity in the responses not found in aroma and basic taste attributes (Data Table is not showed).

The attribute green was unresponsive to all sources of variation. The type of starch or interaction with starch type sources of variation were significant for the attributes milk, starch, cream, and whey. The concentration of starch or interactions with the concentration of starch was significant for starch, caproic acid, plastic, and rancid. Of the 17 attributes,

only green and caproic acid were not significantly affected by days of storage, and the response to days of storage was always a quadratic effect. The attributes baby vomit, potato, valeric acid, and plastic have low intensities (<1) for the duration of the study, and the attributes cooked, powder milk, and cream have initial values greater than 1, but by day 21 they are less than 1. The difference that the type of starch makes for milk and cream is simply additive. The lines for each starch are parallel for the attribute, with corn starch slightly greater in value than kudzu starch. The attributes cream cheese, potato, buttermilk, powdered milk, cooked, cream, plastic, baby vomit, and valeric acid had similar quadratic responses and maintained low intensities throughout storage. Milk, whey with kudzu starch, and rancid flavors decreased linearly over time (Data Figure is not showed). Sour cream, cream cheese, buttermilk, powdered milk, cooked, cream, plastic, baby vomit, and whey also were closely related to the same effects, as each of these flavors contain some of the same flavors and volatile compounds, thus explaining the high correlation. Cream cheese, powdered milk, and cooked flavors also were closely related by possessing some identical and volatile compounds. The starch flavor can be a result of an interaction between the starch in the yogurt formulation and the acids formed during yogurt storage time (Data Table is not showed). The treatments, storage time, and total solids combined with the thickeners significantly affected the flavor and other properties, such as syneresis, texture, acidity, and color of the yogurt samples investigated by Mohammad (2004).

Thickeners and their amounts, as well as storage time, have been found to have a significant effect on the physical, chemical, textural, and sensory properties of strained yogurts (Yazici and Akgun 2004; Makanjuola, 2012). Using different stabilizers gives different flavors because there is a different interaction of various types of thickeners with the yogurt matrix (Routray and Mishra, 2011). Among the 7 different stabilizers used by Mohammad (2004) – which were pectin, guar gum, CMC, carrageenan, sodium alginate, corn starch, and gelatin at 0.4% levels in buffalo milk with 16.6% total solids, in cow milk with 13.5% total solids, and in a mixture (1:1) of both having 15.0% total solids – the best result in terms of flavor was obtained with a combination of 0.4% corn starch and 16.6% total solids. The influence of the gel structure on flavor release was also observed in this case and was found to be in agreement with sensory characteristics previously studied for these products.

Conclusion

Overall, the addition of kudzu or corn starch yielded very similar responses for most sensory attributes. The ultimate emphasis in yogurt production is placed on taste and quality. Plain yogurt with added kudzu or corn starch has a potential to succeed in the marketplace. Use of kudzu starch may provide a smooth and bright appearance and good flavor in fortified yogurt.

It is evident from this study that yogurt can be produced with kudzu starch at appropriate concentrations (2%). Therefore, the production of yogurt with kudzu starch, which may be consumed because of the availability of kudzu starch in commercial quantities, is another way of increasing the food value of the crop. Processors would need to consider how the products are processed, which could affect the sensory properties of finished products and how consumers will perceive them. Further study is necessary to examine the rheological measurements of yogurt samples during storage period.

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