COMPARISON OF BREAD KVASS FERMENTED WITH DIFFERENT YEASTS

[a]IVO LIDUMS, [b]DAINA KARKLINA, [c]ASNATE KIRSE

Department of Food Technology, Faculty of Food Technology, Latvia University of Agriculture, 22 Rigas Street, Jelgava, LV3004, Latvia
email: [a]ivo@ilm.lv, [b]daina.karklina@llu.lv, [c]asnate.kirse@llu.lv

Abstract: The aim of this study was to compare physicochemical and sensory characteristics of kvass fermented with *Saccharomyces cerevisiae* and *Kluyveromyces marxianus*. Naturally fermented bread kvass was made from dried rye bread rusks. Kvass fermented with *S. cerevisiae* had higher dry matter content (8.6%) and lower pH (3.88) compared to kvass fermented with *K. marxianus* (7.0% and 4.60, respectively). Results of hedonic evaluation did not show significant differences ($p>0.05$) between the preference of kvass samples among panelists; consumers, however, preferred kvass fermented with *S. cerevisiae*. Evaluators reported considerable differences in colour ($p<0.05$) while aroma of both kvass samples was similar.

Keywords: kvass, non-traditional yeast, hedonic evaluation, *Saccharomyces cerevisiae*, *Kluyveromyces marxianus*

1 Introduction

1.1 Kvass beverage

The past several decades have seen a sharp increase across the globe in the consumption of caloric beverages [Popkin, 2012]. Although their regular intake has been associated with human health problems, carbonated soft drinks remain one of the most frequently consumed ready-to-drink beverages in the world [Francisco et al., 2015]. Soft drinks are defined as water-based flavoured drinks usually with added carbon dioxide and with nutritive, non-nutritive, and/or sweeteners with other permitted food additives [Ryan, 2014]. They present particular characteristics such as high sugar content and acidity, and are available on the market in many flavours [Francisco et al., 2015].

Kvass is a soft drink traditional to Eastern European countries, typically produced by fermenting kvass mash with yeast (traditionally *Saccharomyces cerevisiae*); alcohol content in kvass must be less than 1.2% alcohol by volume. Kvass, a non-alcoholic beverage, is produced from rye or dried rye bread by natural fermentation [Lidums et al., 2014]. Kvass quenches thirst, is invigorating and refreshing. Because of the favourable composition of the microflora (lactic acid bacteria, yeast), kvass is enriched with B group vitamins, as well as lactic acid and carbon dioxide; it is a product of incomplete alcoholic and lactic acid fermentation [Omasheva et al., 2015]. Kvass has beneficial effects on the digestive tract; furthermore, energy value of naturally fermented kvass is only 25 kcal (105 kJ) per 100 ml [Lidums et al., 2016], which is about ½ less than in typical soft drinks. Most of the beneficial substances come from the raw materials used in naturally fermented kvass production – rye bread and malt, especially in non-pasteurized kvass.

1.2 Yeast alternatives

There is tremendous diversity among yeasts and the application of modern microbiological and molecular approaches has resulted in renewed focus on the biology and industrial potential of yeasts other than *S. cerevisiae*. Nowadays, an upsurge of interest in providing suitable starter and adjunct cultures for food production has been observed. Many researchers have proposed a variety of cultures as alternatives to baker's yeast, including *Kluyveromyces marxianus* [Plessas et al., 2008]. The dairy yeast *K. marxianus,* described as a homothallic, hemiascomycetous yeast, is phylogenetically related to *S. cerevisiae* and positive for assimilation a wide range of sugars [Lane, Morrissey, 2010]. As a dairy yeast, K. *marxianus* can be grown in cheese whey (lactose fermentation), which is the main by-product of the dairy industry, and its disposal causes serious environmental problem [John et al., 2007]. K. *marxianus* has also been successfully used in bread making instead of baker's yeast [Dimitrellou et al., 2009] and for agave must fermentation in tequila production, showing increased volatile compound content comparing with a *S. cerevisiae* baker's yeast [Lopez-Alvarez et al., 2012].

Recently, a *K. marxianus* strain has been experimentally grown and studied in a non-conventional dairy product factory in Latvia, therefore additional uses of this yeast in other food sectors have been considered. One of the potential uses could be kvass production, as *K. marxianus* is positive for glucose and sucrose fermentation.

Sensory parameters such as aroma, taste and colour are critical in the acceptance of products; therefore, the aim of this research was to compare physicochemical and sensory characteristics of kvass fermented with *S. cerevisiae* and *K. marxianus*.

2 Materials and methods

2.1 Microorganisms

Strain *K. marxianus* DSM 5422 was obtained from Leibniz-Institute DSMZ-German Collection of Microorganisms and Cell Cultures and maintained on agar plates containing 2% glucose and recultivated in semi synthetic medium containing lactose 50 g/L, yeast extract 5 g/L, $MgSO_4 \cdot 7H_2O$ 1.4 g/L, KH_2PO_4 1.0 g/L, K_2HPO_4 0.1 g/L, $(NH_4)_2SO_4$ 5.0 g/L at 30℃ with agitation 180 rpm. The wet cells were harvested by centrifugation at 5000 rpm for 10 min and then were used for kvass preparation.

Dry baker's yeast S. *cerevisiae* from Lallemand Ltd. was used for comparison.

2.2 Kvass preparation

For bread kvass production, the additional materials were used: rye bread rusks (Liepkalni Ltd.), lactic acid bacteria *Leuconostoc mesentericus* (Chr. Hansen Ltd.), beet sugar (Dansukker Ltd.) and dark malt (Liepkalni Ltd.).

Kvass was produced according to Lidums et al. [2016]: for 1 litre of kvass mash, 200 g of rye bread rusks and 2 g dark malt were soaked in 2 litres of hot water (78±2 °C). Bread rusks were left to soak for 3 hours, then the water-bread rusk suspension was filtered (300 microns) and the liquid fraction was cooled down. 1 g baker's yeast (or proportionate amount of wet *K. marxianus*), 2 units of lactic acid starter and 30 % of the estimated quantity of sugar were added to 1 litre of kvass mash. The total quantity of sugar for kvass production is 30 g; therefore 10 g of sugar were added prior to fermentation. The fermentation of kvass mash took 9 hours at 29 ± 1 °C. After fermentation kvass was placed in a refrigeration chamber to cool down to 3 ± 1 °C. After cooling, the remaining sugar was added (blending). Kvass was maturated for 12 hours at 6 ± 1 °C (total production time 25 hours). Afterwards kvass was filled in 0.5 l PET bottles and stored at 4 ± 1 °C for 12 hours before physicochemical analysis and sensory evaluation.

2.3 Physicochemical analysis

Kvass quality parameters are defined by the Regulation No 926/2010 "Quality and classification requirements for kvass and kvass (malt) beverage" of the Cabinet of Ministers of the Republic of Latvia. The parameters defined are 1) dry matter content - 3.0 to 14.0 percent by weight, and 2) acidity - 2.0 to 3.5, expressed as ml of 1n NaOH per 100 ml. Active acidity (pH) of kvass samples was measured by a pH meter (Jenway 3510, Jenway, UK) and dry matter (Brix°) by a digital refractometer (DR301-95, Krüss, Germany).

2.4 Sensory evaluation

A total of 30 trained panellists (18 men and 12 women, average age 22 years) from Faculty of Food Technology, Latvia University of Agriculture participated in initial testing of kvass fermented with *S. cerevisiae* (sample A) and *K. marxianus* (sample B).

As views of consumers are very important, 150 consumers (57% women and 43% men, average age group 25-45) were asked to complete a short questionnaire about their kvass consumption habits and participate in kvass tasting during the Baltics food industry fair ‘Riga Food 2015’. 5-point hedonic evaluation (5 – like very much and 1 – dislike very much) was used in order to determine the overall preference of the samples, while line scale showed the intensity of kvass sensory properties (aroma, flavour, acidity, and colour) [Lawless, Heymann, 2010].

2.5 Data analysis

The obtained data processing was performed using mathematical and statistical methods with Microsoft Excel v16.0; differences among results were considered significant if p-value < 0.05. One-way analysis of variance (ANOVA), Tukey’s test and independent samples *t*-test were used.

3 Results and discussion

3.1 Physicochemical parameters

Dry matter content in sample A (kvass fermented with *S. cerevisiae*) was 8.6 ± 0.1% and in sample B (kvass fermented with *K. marxianus*) – 7.0 ± 0.1%. Active acidity (pH) was lower in sample A (3.88 ± 0.02) compared to sample B (4.60 ± 0.03). According to the indexed values of the Regulation No 926/2010, dry matter content and pH value of laboratory produced kvass corresponded to the regulatory scale (experimentally determined conversion factor for pH ~1.8).

3.2 Kvass consumption patterns

Consumption of kvass in mainly dependent on the weather outside, as majority of women (52%) and men (27%) drink kvass when the weather is warmer, i.e., during summer months (Fig. 1.).

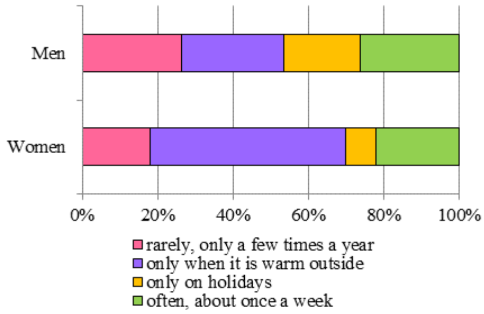

Figure 1: Frequency of kvass consumption

Approximately ¼ of both genders consume kvass once a week and about the same amount of consumers rarely drink kvass. For some men (20%) holidays are the time when they increase kvass consumption. There is a similar amount of consumers of both genders who consume kvass frequently or drink it rarely.

3.2 Kvass sensory evaluation

According to trained panellists the overall preference of kvass drinks range from 4.1 – ‘like a little’ to 5.0 – ‘like very much’ (Fig. 2). Hedonic evaluation showed that there were not significant differences (p>0.05) between the preference of kvass samples among panellists. Results of consumer evaluation reported considerable preference to kvass sample A (p<0.05), which was within the range from ‘like a little’ to ‘like very much’. Preference of kvass sample B ranged from 3.5 – ‘not sure’ to 4.0 – ‘like a little’. Preference of kvass fermented with *S. cerevisiae* (sample A) between panellists and consumers was similar (p>0.05), yet consumers liked fermented with *K. marxianus* (sample B) less than panellists (p<0.05).

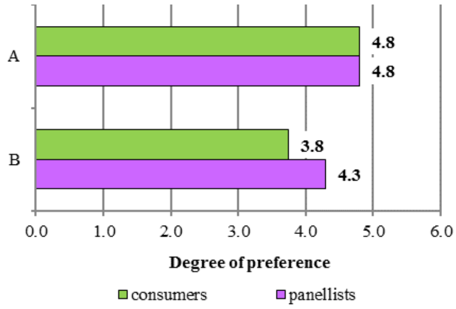

Figure 2: Results of hedonic evaluation of kvass samples

The majority of consumers (56%) reported they would probably not drink kvass with *K. marxianus*, while 15% would not drink kvass with *S. cerevisiae*. 60% of consumers would like to consume kvass with *S. cerevisiae* regularly as it has a more traditional taste. Some consumers liked the aroma, acidity and aftertaste in kvass with *S. cerevisiae*; however others pointed out that it did not have enough acidity, bitterness and CO_2. Kvass with *K. marxianus* was reported to have not enough acidity and aroma; it was too light for typical kvass. Children (of consumers) who did not participate in sensory evaluation said that this sample tasted like lemonade.

This brings us to consider soft drink consumption patterns of consumers, as most of the commercially available beverages sold as kvass are kvass drinks made by diluting grain extract concentrates with water and adding colourings, flavours and artificial sweeteners. Kvass drinks are sometimes produced without the use of yeast, therefore carbon dioxide is added artificially for no fermentation has taken place [Lidums et al., 2015]. Naturally fermented kvass is less likely to have the amount of carbon dioxide found in soft drinks with added CO_2, as yeast produces lower amounts of longer lasting carbon dioxide during natural fermentation. Therefore, naturally fermented kvass could be preferred less than its rival kvass drinks with high CO_2 content, if consumers prefer highly carbonated beverages.

In order to evaluate and compare the intensity of kvass sensory properties, line scale was used. Consumer evaluation showed significant differences between kvass sample A and B in terms of flavour and colour (p<0.03) however aroma and acidity of kvass samples was similar (p=0.33) (Fig. 3.).

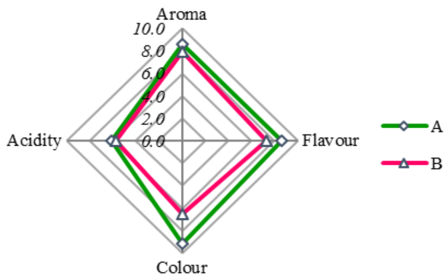

Figure 3: Consumer evaluation of the intensity of sensory properties of kvass samples

Panellists reported considerable differences between colour and acidity of kvass samples (p<0.03), but aroma and flavour were comparable (p>0.26) (Fig. 4).

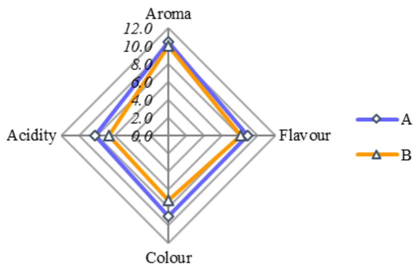

Figure 4: Panellist evaluation of the intensity of sensory properties of kvass samples

Consumers reported that kvass fermented with *S. cerevisiae* (sample A) had a more pronounced flavour while panellists found similar flavour in both samples. Dimitrellou et al. [2009] pointed out that analysis of volatile aroma-related compounds and the amounts of total volatiles in bread samples (breads produced by baker's yeast and K. *marxianus*) were not significantly different. Panellists, on the other hand, noted higher acidity in kvass sample A that is also shown as lower pH value.

Colour intensity differences were reported by both groups of evaluators. Kvass sample A had a darker, more intense colour than sample B. Food colour usually is the first quality parameter evaluated by consumers which is critical in the acceptance of products [Cserhalmi et al., 2006]. However, there are no regulations for kvass colour as it is dependent on ingredients used; it is possible that there were inconsistencies within the colour of rye bread rusks and dark malt, therefore, resulting in various colour intensities of kvass mash.

When comparing intensity of sensory properties evaluated by consumers and panellists of the same kvass sample, sample A showed differences between aroma and acidity intensity (Fig. 6).

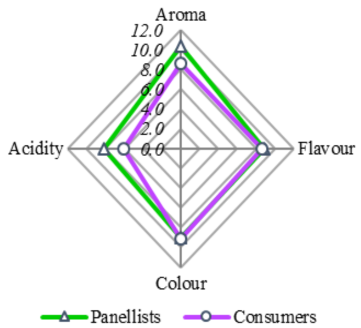

Figure 5: The intensity of sensory properties of kvass fermented with *S. cerevisiae*

Panellists reported more pronounced aroma and acidity of kvass sample A ($p<0.01$), while flavour and colour had insignificant differences among evaluators. As for kvass sample B (Fig. 6), panellists found kvass aroma to be significantly more intense ($p<0.01$) contrary to consumers. Panellists noted that acidity, flavour and colour of kvass sample B was more distinct, however there was only borderline significance ($p<0.1$).

Consumer soft drink consumption habits play a great role in preference of kvass samples and intensity evaluation of sensory properties. Differences in consumer evaluation are always linked with psychological and environmental aspects [Lawless, Heymann, 2010].

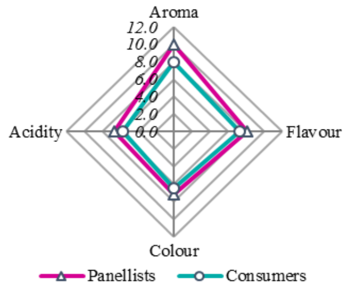

Figure 6: The intensity of sensory properties of kvass fermented with *K. marxianus*

It should be noted that sensory properties of kvass also depend on technological process; therefore kvass produced at a factory could have diverse intensity of sensory properties contrary to kvass produced at laboratory.

4 Conclusions

Dry matter content and active acidity of kvass samples was within the admissible range according to Regulation No 926/2010 of the Cabinet of Ministers of the Republic of Latvia. Consumer consumption of kvass is mainly dependent on the weather outside, as majority of drink kvass when the weather is warmer, while approximately ¼ of both genders consume kvass once a week. Hedonic evaluation showed that there were not significant differences ($p>0.05$) between the preference of kvass samples among panellists. Preference of kvass fermented with *S. cerevisiae* between panellists and consumers was similar ($p>0.05$), yet consumers liked fermented with *K. marxianus* less than panellists ($p<0.05$). Consumer evaluation showed more pronounced flavour and colour in kvass fermented with *S. cerevisiae*, but aroma and acidity of kvass samples was similar. Panellists reported more pronounced colour and acidity in kvass fermented with *S. cerevisiae*, yet aroma and flavour were comparable. The results suggest that *K. marxianus* DSM 5422 could be suitable for kvass fermentation and production; however additional evaluation of kvass produced at a factory should be carried out.

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