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ORIGINAL RESEARCH PAPER

RESEARCH ON THE USE OF DEPROTEINISED WHEY IN BEER PRODUCTION

Elena R. Huber[1], Adriana Dabija[2], Ancuța Chetrariu[2]*

[1]Stefan cel Mare University of Suceava, Doctoral School of Applied Sciences and Engineering, 720229 Suceava, Romania
[2]Stefan cel Mare University of Suceava, Faculty of Food Engineering, 720229 Suceava, Romania

*Corresponding author: ancuta.chetrariu@fia.usv.ro

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Abstract: Preventing waste generation is the top priority of the New Waste Framework Directive (Directive 2008/98/EC), which then places the least emphasis on disposal and more on processing for reuse (valorization), recycling, and recovery. In a similar vein, the European Union's bio-economy policy needs to be implemented in order to enhance biological resource management, create new markets for food and biobased goods, and protect the environment. Dairy processors must therefore create efficient and lucrative methods of managing whey waste, especially given the exorbitant prices of the waste treatments used today. The consumers, business, and scientific community are becoming more interested in the recycling and reuse of food and agricultural industry by-products. Deriving added value from otherwise lost food production outputs is a key principle of the circular bioeconomy, even though there is still disagreement over a common definition of this notion. Whey, a by-product generated during the manufacture of cheese and casein-based dairy products, is rich in nutrients and has several commercial uses. This by-product can be used in various value-added products with the help of advanced technologies. Obtaining whey beverages represents a viable solution for valorization that can be used by cheese producers in our country. The paper proposed the valorization of deproteinised whey in the manufacture of whey beer. It was used in a proportion of 30 % as a substitute for water in the beer production process. Also, to improve the physical-chemical and sensory characteristics, local syrups were used. The most appreciated variant from a sensory point of view was the whey beer with fir bud syrup. The finished product - whey beer, is a product with properties similar to beer: pleasant, characteristic smell, with hop aroma, pleasant bittersweet taste.

Keywords: *by-product, circular bioeconomy, food waste, valorization, whey beer*

INTRODUCTION

Worldwide, about half of milk production is processed into cheese, and up to 90 % of the milk volume is found in whey, which generates a massive secondary flow with a high organic load and potential environmental impact if not managed correctly [1]. One way to valorize whey is to obtain cheese from whey. This technological process results in deproteinized whey, which represents about 90 % of the initial whey. It retains up to about 6 % lactose from the milk composition and a significant amount of mineral substances, but has levels of biochemical oxygen demand (BOD) and chemical oxygen demand (COD) of about 50 $g \cdot L^{-1}$ and 80 $g \cdot L^{-1}$, respectively, well above acceptable limits for urban wastewater, being classified as waste if not reused as raw material, direct discharge being prohibited [2]. Therefore, the disposal of deproteinized whey represents a problem for cheese production units, many of which opt to use it as animal feed [3].
A way of valorizing this waste into a particularly valuable by-product, also proposed in this work, is to obtain deproteinized whey beverages, being one of the simplest and most profitable options for using whey in human nutrition. It has also been found that combining it with fruit juices is an interesting option for improving the sensory quality of whey beverages [4]. Whey beverages represent an innovative method of using whey for human consumption, both in the form of unfermented and fermented beverages, which can be enriched with nutraceutical, probiotic and prebiotic components and can be combined with fruit juices or milk [1, 3]. In practice, safe formulations maintain *p*H below 4.6, and the addition of fruit and vegetable juices increases acidity, reduces viscosity and improves sensory perception [5, 6]. They can be functional beverages with a “health & sustainability” positioning, and on-site processing in small units reduces waste and transport costs. Major limitations are related to high perish ability, variability of composition depending on species, feed, season and technology, as well as the presence of lactose, which requires either the use of lactase or an appropriate fermentation, along with the need for standardization of raw materials and rigorous sensory validations [3]. From a circular economy perspective, the valorization of deproteinized whey moves purification costs into food/functional added value, aligning with the European Green Deal and trends in “functional beverages” [5, 7].
One of the beverages for which deproteinized whey has been experimented with is beer, which is one of the most widely consumed low-alcohol fermented beverages in the world [8 – 11]. In recent decades, the increase in beer production has led to the production of both traditional and new types of beer [12]. New unconventional approaches also include locally sourced ingredients, fruits, spices, vegetables, and other natural ingredients that can be used in manufacturing recipes, and are also considered economical and healthy ways to obtain unique characteristics of the finished product [8, 13, 14]. Fruits are one of the most common ingredients added in the brewing process [15]. They can add new compounds, with influences on the physicochemical and nutraceutical properties, for example, specific polyphenols, and in this way can increase the concentration of bioactive compounds in the finished beer [16]. When obtaining some types of beer, the addition of berries, oranges, pineapples, apples, apricots, plums, cherries, raspberries, seabuckthorn, rosehips, etc. has been experimented with [15, 17, 18]. For example, Mileva *et al.* (2018) added 100 $g \cdot L^{-1}$ of blueberries during beer maturation and found a 1.3 to 1.4-fold increase in polyphenols and a 1.2 to 1.4-fold increase in flavonoids [19].

The beer industry is trying out cutting-edge ways to increase its overall sustainability, one example of such practices being the use of local ingredients in beer brewing. Few studies have explored consumer and producer interest in beers made with local fruits or agro-industrial by-products [20]. Some of these innovations have focused on the use of local ingredients, which add value to products, reduce production costs and environmental impact, and provide beers with special organoleptic characteristics [7, 21 – 23].
The paper presents its own research in this direction of capitalizing on deproteinized whey and some local ingredients (raspberry, elderflower, fir buds) in the classic manufacturing recipe for obtaining blonde beer.

MATERIALS AND METHODS

Production and analysis of the samples were carried out in the Stefan cel Mare University of Suceava, Faculty of Food Engineering.

Raw materials

Malt, hops and yeast were purchased from the Bermas SA Suceava brewery. In the experiments, sweet deproteinized whey resulting from the manufacture of whey cheese (in Romanian ”*urda*”) from the company Five Continents Group SRL Romania was used with the following characteristics: total dry substance - 5.48 %, of which, fat - 0.44 %, proteins - 0.43 %, lactose - 4.50 %, acidity - 18.2 °T (Thörner acidity degrees), density - 1.0195 $g \cdot L^{-1}$. The raspberry syrup, elderflower syrup and fir buds syrup was purchased from a local processing company, Nord Natural Hub SRL (Campulung Moldovenesc, Suceava County). Also, to obtain the beer in laboratory conditions, hops from the Hallertau Blanc Company (α-acids content 10.1 % by weight (w/w)) and Fermentis brewer's yeast type 74/30 were used.

Beer production

The beer wort was obtained according to a conventional method of obtaining beer with following main operations: mashing, filtration, boiling and cooling. In the manufacturing recipe, 30 % of the water was replaced with deproteinized whey. The beer wort obtained presented the following characteristics: extract content 11.7 % w/w, *p*H = 5.7, color = 0.44 EBC, bitterness value (IBU) = 7.54 EBC, density = 1.0411. For sensory improvement, the following types of syrup were introduced into the hopped wort, before the technological operation of primary fermentation, in a proportion of 5 %: raspberry (S1), elderflower (S2) and fir buds (S3). The addition of these syrups led to an increase in the extract content of the must before fermentation to a value of 13 % w/w. The control sample was the beer sample without added syrup.
After cooling to 12 °C, the wort was inoculated with 25 $mL \cdot L^{-1}$ of yeast type 74/30 Fermentis, the quality of which was previously determined using the Nucleocounter YC-100 (ChemoMetec A/S, Denmark), the number of total cells being 20 x 10^8 cells $\cdot mL^{-1}$ biomass, of which dead cells 1.28 %. The primary fermentation took place at a temperature of 12 °C for 6 days, in an industrial refrigerator with the possibility of digital regulation. Then, after cooling down to 2 °C, the yeast was discharged from the

bottom of the vessel. The maturation lasted for 20 days at 4 °C in the same refrigerator industrial.

Beer analysis

At the end of maturation, the beer was analyzed from the sensorial and physicochemical point of view. All analyses of beer technological parameters were carried out in accordance with recognized methods within European Brewery Convention (EBC Analytic, 2010): the real and apparent extract (% w/w; EBC 9.4), the alcohol content (% v/ 164 v, % w/w; EBC 9.2.1), density ($g \cdot cm^{-3}$; EBC 9.43.1), turbidity (EBC 9.43.2), *p*H (EBC 9.35), color according to EBC 9.6, bitterness value (IBU, 166 EBC 9.8), CO_2 content (EBC 9.28.1), O_2 content (EBC 9.37.1), calories content kJ/100 mL (EBC 9.45) [24]. These methods can be applied to all types of beers. Three determinations from the same sample were performed in parallel for analysis and the mean value was used in this study.
The color of the beer samples was evaluated and with a Konica Minolta CR-400 colorimeter (Tokyo, Japan), using CIELab color space coordinates, where L* values describe black (0 value) to white (100 value), a* is the degree of redness (positive) or greenness (negative), and b* is yellowness (positive) or blueness (negative). Samples were measured in triplicate and a white standard was used for calibration [25].

Sensory analysis

For sensory analysis, the scoring scale method was used, a standard method in Romania. The method consists of evaluating each organoleptic property, by comparing with scoring scales of 0 ÷ 5 points and obtaining the average score of the group of tasters, calculating the weighted average scores, summing them, to obtain the total average score and establishing the organoleptic quality of the product, based on the total average score, by comparing with a scale of 0 ÷ 20 points (SR 13355-1/June 1997). A trained panel consisting of 15 members conducted the sensory evaluation of the samples. The test was performed in the Sensory Analysis Laboratory of the Stefan cel Mare University of Suceava, Faculty of Food Engineering. Approximately 70 mL of beer sample coded with three-digit numbers were served to the panellists in random order at 8 °C in glass cups. Between samples the panellists used unsalted crackers and water to cleanse their mouths to reduce taste transfer from one sample to another [26].

RESULTS AND DISCUSSIONS

Physicochemical characteristics of whey beer

The variants of the finished product have been developed to optimize sensory and physical-chemical characteristics. The whey beers obtained by the three variants differed from the control sample in terms of real extract, apparent extract, alcohol content, carbon dioxide content, while insignificant differences were observed in color, *p*H, and bitterness value. A very important distinguishing parameter is ethyl alcohol, the content of which was affected using of whey and added syrups. The results of the beer samples taken into study are summarized in Table 1.

Table 1. *Physicochemical characteristics of whey beer-finished product*

Characteristics	Sample			
	Control sample [CS]	S1	S2	S3
Real extract [% w/w]	5.81±0.4	9.54±0.05	10.04±0.04	10.56±0.05
Apparent extract [% w/w]	3.77±0.02	7.73±0.07	7.95±0.06	7.98±0.06
Alcohol content [% v/v]	5.20±0.06	4.90±0.09	4.80±0.05	4.70±0.04
Alcohol content [% w/w]	4.08±0.05	3.74±0.08	3.68±0.05	3.58±0.04
Density [$g\cdot cm^{-3}$]	1.0136±0.01	1.0288±0.03	1.0379±0.02	1.0437±0.02
Turbidity [EBC units]	0.86±0.02	3.19±0.04	3.03±0.03	1.98±0.01
*p*H	4.65±0.03	4.63±0.09	4.14±0.05	4.16±0.05
Color [EBC units]	9.00±0.10	11.77±0.18	6.11±0.16	8.00±0.11
Bitter value [IBU units]	25.8±0.53	24.9±0.95	23.8±0.60	24.8±0.69
CO_2 [$g\cdot L^{-1}$]	5.12±0.07	4.42±0.03	4.42±0.01	4.46±0.01

From Table 1 it can be seen that all samples with added syrup have much higher values of real extract content and apparent extract content than the control sample. This is due to the contribution of fermentable carbohydrates present in the syrups, which led to initial extract content higher than that of the control sample (13 % w/w). It is also known that lactose is a non-fermentable disaccharide that can influence, in addition to the sensory properties, the extract content of the finished product. The increased real and apparent extract and the associated increase in density in the syrup variants are not unexpected, as many research claim that added sugars in syrup and lactose increase residual extract and density in beers due to unfermented carbohydrates [27]. From Figure 1 it can be seen that, compared to the control sample, the values of the real extract and apparent extract content in the beer samples with added syrups are much higher, due to the amount of non-fermentable carbohydrates contained in them.

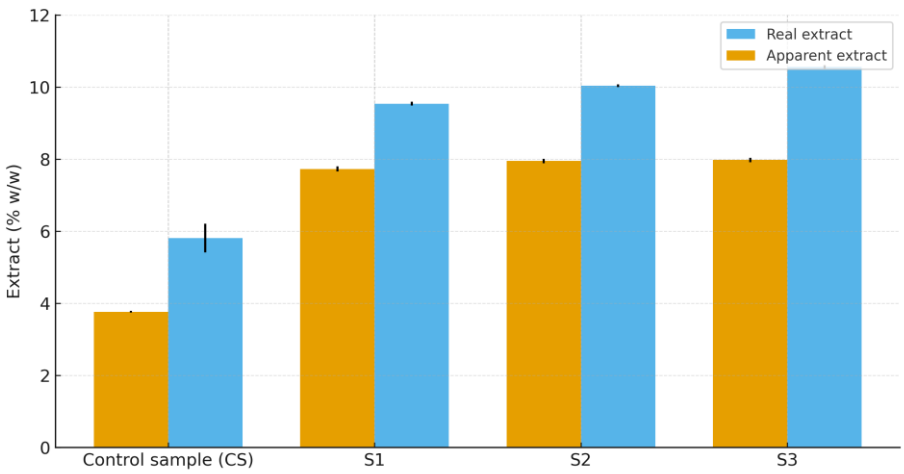

Figure 1. *Real vs. apparent extract content in all whey beer samples*

The ethyl alcohol content for all samples with added syrup is lower than the control sample. This is a consequence of the presence of lactose and non-fermentable compounds in the syrups, which limit the conversion to alcohol. On the other hand, the increase in density in all beer samples confirms the high extract content and the presence of residual compounds in the finished product. The reduction in alcohol content of the syrup-added whey beers is consistent with other studies showing a reduction in alcohol content due to the presence of lactose and non-fermentable sugars in the syrup, which limits fermentation and therefore alcohol production. The literature on fruit additions to beer similarly document that fruit syrups may dilute the beer or add non-fermentable sugars and reduce or have negligible effects on the alcohol content of the final product, supporting the current findings [28].

The addition of syrups led to an increase in turbidity, the highest values were recorded by the beer with added raspberry syrup and elderberry syrup, respectively 3.19 units EBC and 3.03 units EBC. Regarding the *p*H, the values are close to the control sample, the lowest values being recorded by the beer with added elderberry syrup (*p*H = 4.14), followed by the one with added fir bud syrup (*p*H = 4.16), probably due to the organic acids in the respective syrups. The slight decrease in *p*H for elderberry and fir buds syrup beers reflects organic acids associated with fruit syrups, a trend seen in other fermented beverages based on whey and fruit [29]. Also valuable in craft fruit beer studies, is the recognition of the effects of sweetness and aroma compounds in syrup, which can mitigate the bitterness from hop compounds [29].

The bitterness value recorded similar values for all beer samples, the differences compared to the control sample are explained by the attenuation of the bitterness value brought by the hops by the sweet and aromatic compounds of the syrups used in the beer manufacturing recipe.

The CO_2 content of the three samples with added syrup was lower than that of the control sample, in the samples with raspberry syrup and elderberry syrup, by 13.67 %, and in the beer sample with fir bud syrup by 12.89 % compared to the control sample. In conclusion, all three samples with added syrup have high extract content, high density, but a lower alcohol and carbon dioxide content. Greater differences are recorded in the beer with raspberry syrup, which has a more intense color and high turbidity, while the beer with elderberry syrup has a higher *p*H, but the lightest color. From the point of view of physico-chemical characteristics, the most balanced and closest to the control sample is the beer with fir buds syrup.

To highlight the differences in color of beer samples with different syrup additions, color analysis was also performed using the colorimetric method [25]. The results obtained are summarized in Table 2.

Table 2. *Color of whey beer-finished product*

Sample of whey beer	**L**	**a***	**b***
Control sample	29.54	0.04	9.07
S1	25.97	5.16	5.79
S2	27.98	-0.29	10.17
S3	28.18	-0.49	3.45

Whey beer samples vary in L*, a*, b* values, which is due to the different syrups used for their production. S1 have lower L* values and high a* values, which mean that they

have a darker reddish color. This is due to the anthocyanins, which are the polyphenols responsible for the red color of fruit. Samples S2 and control sample have their b* values similar, which mean that they are more yellowish than S1 and S3. The highest value of a* is for S1 sample, which mean that is the redness whey beer sample and the lowest a* value is for S3 sample, which means is the greenness sample. The differences in color parameters (L, a, b) are congruent with specified effects of polyphenols, for example, anthocyanins from fruit syrups produce red-brown colors and higher turbidity [30]. Literature indicates that fruit beers usually have greater turbidity and darker or more intensely colored profiles and it depends on type of fruit and concentration, which is congruent with the raspberry syrup our sample's characteristics [31].
The physicochemical properties recorded for the examined whey beers are relatively consistent with previous literature, confirming that the lactose and protein content of whey significantly contribute to the expected effects on extract, alcohol, density, *p*H, color, and turbidity, all consistent with findings from previous studies into whey beers [32]. This indicated the reproducibility and expected role of whey composition on beer properties.

Sensory characteristics of whey beer

The most important aspect of creating products that live up to consumer expectations is sensory analysis. The evaluation of appearance, aroma, taste, texture, and overall quality through standardized procedures involving trained judges or consumer panels is therefore frequently a complement to investigations into the incorporation of health-promoting functional ingredients like syrups into traditional recipes. Analytical workflows that aim to isolate and determine the concentration of volatile compounds in foods and comprehend the impact of food components on the overall intensity of the aroma and/or final product perception are frequently used in conjunction with the sensory evaluation of aroma, which is particularly important in stimulating consumer preferences [26].
Figure 2 shows the four beer samples that were analyzed by the tasters. From the analysis of the results presented in Table 3, it is observed that the highest score was presented by S3, the whey beer with added fir syrup, which received the rating of „very pleasant beer”. The other two samples, whey beer with added raspberry syrup and beer with added elderberry syrup received the rating of „pleasant beer”. Whey beer presents distinct sensory, functional and ecological characteristics, located at the intersection of food innovation, the valorization of renewable resources and the circular economy. With an original recipe, it presents special sensory characteristics, a unique flavor, a bittersweet taste, with citrus nuances and unmistakable freshness.

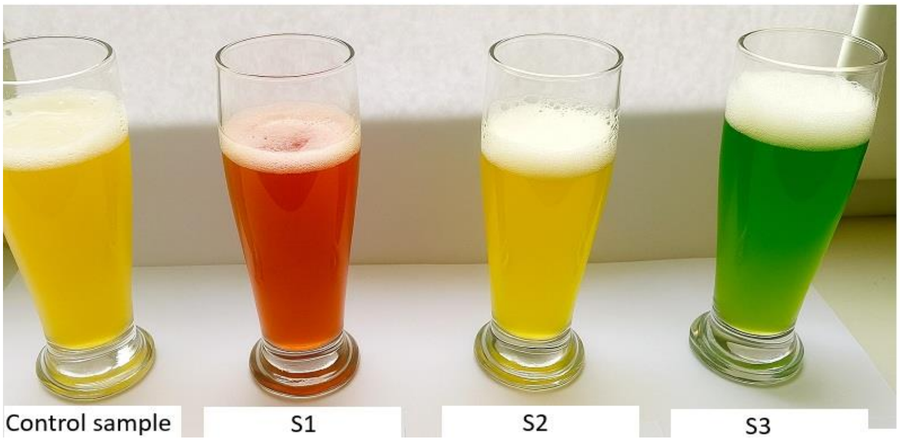

Figure 2. *Whey beer samples: Control sample (whey beer); S1- whey beer with raspberry syrup; S2 - whey beer with elderberry syrup; S3 - whey beer with fir buds syrup*

Table 3. *Synthesis of sensory analysis results for whey beer samples*

Sample analyzed	Average total score	Qualifying	Product characterization
CS	19.24	Very pleasant beer	The beer has positive, specific organoleptic properties that are well defined. It does not present any type of perceptible deficiencies or defects.
S3	19.50		
S1	17.56	Pleasant beer	Beer has positive, specific, quite defined qualities, but also very small, insignificant shortcomings and defects.
S2	17.82		

The sensory analysis findings further confirm information from other research relevant to whey based, fruit flavored, fermented beverages. More general, the combination of standardized sensory evaluation by trained judges or consumer panels and analytical consequence of volatile compounds and overall aroma perception is considered to be essential when examining product acceptance and consumer preferences for these functional beverages [33].
The highest sensory score went to the whey beer with fir buds syrup (S3) which was labelled "very pleasant beer," while wines with raspberry syrup (S1) and elderberry syrup (S2) ranked slightly lower, but still quite pleasantly met the consumer expectations, is consistent with the well-established belief that syrup additions can modify sensory and generally improved sensory acceptability [34]. Pure whey is often poorly accepted but fruit juices or flavors added to whey lessen the watery cheese taste and aroma, and are potential solutions to support consumer liking.
While it is recognized that whey beers have unique sensory properties related to bitter-sweet taste, unique flavors, and freshness; there are other studies that contend whey can provide its own sensory property characteristics when used as an ingredient in beer, considering and optimizing brewing conditions [34]. Whey in general can have sour, aftertaste in addition to salty; with a high salt mineral content and whatsoever it can contribute to unpalatable flavor. Regardless, this evidence contributed to the unique final beverage profile.
The presentation of sensory analysis is a standard, complementary method of sensory evaluation on novel beer formulations [33]. Sensory analysis is something that requires

serious attention as aroma is one the prime and overall quality traits that consumers look at and indicate for quality issues pertaining to time, storage, contamination, or raw materials to support or validate product quality.

Moreover, as found in the literature, our research confirms findings more broadly from sensory analysis that syrups have the potential to promote sensory qualities of whey beer specifically, and that rigorous sensory evaluation will be necessary with respect product development and product acceptance by consumers [34]. Creating new products that have new and tasty characteristics is a substantial trend in the food industry, driven by consumer demands for healthier functional beverages that also appeal to taste.

CONCLUSIONS

Whey alcoholic beverages are suitable for a broader range of consumers, primarily due to their low lactose content and reduced allergenicity. Moreover, compared to non-alcoholic whey drinks, whey alcoholic beverages offer extended shelf life, higher levels of phenolic compounds, and enhanced antioxidant activity. Whey beer, the final result, has characteristics comparable to those of beer, including a nice, distinctive smell, a hop aroma, and a pleasant, bittersweet taste. The sensory attributes of the whey beer product, like its unique aroma of hops and the taste characteristics as bittersweet with the added color attributes of the green color from the colored syrup of fir buds, makes it unique in the market of beverages. The sensory attributes of this beverage will enhance the consumer's experience and possibly cater to a market beyond traditional beer drinkers.

With regards to production, whey beer can be produced on an industrial level with relatively simple processes, and equipment that could involve existing brewery equipment and processes. The ability to promote process scaling so that it can be produced from a microbrewery, to a vastly larger capacity facility while adhering to food safety laws and standards of quality, makes the industrial innovation meaningful. The methods of production are relatively simple and flexible which makes it easy to pivot for cost effective and reproducible processes in the market, to consumers. Therefore, whey alcoholic beverages represent the functional and sensory innovation, as well as offering relevance for broad industrial application with an economic and environmental potential of valorizing dairy by-products. Thus, the study showed how a low-alcohol, whey-based beer can be made at an industrial level that meets the needs of regulation, food safety and consumer desires, creating a need in the market for inclusion into a consumer driven market for functional and specialty drinks.

In conclusion, the project proposes obtaining a non-distilled low-alcoholic beer-type beverage that can be easily reproduced on an industrial scale, from micro-factories to large-capacity production companies, through a simple process, in accordance with the provisions of the regulations in force, under food safety conditions.

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