Scientific Papers. Series B, Horticulture. Vol. LXVI, No. 1, 2022
Print ISSN 2285-5653, CD-ROM ISSN 2285-5661, Online ISSN 2286-1580, ISSN-L 2285-5653

# REVIEW: ACTUAL APPROACHES FOR THE CRAFT BEER FERMENTATIONS

**Carmen-Rodica VRÎNCEANU[1], Iuliana Diana BĂRBULESCU[1], Florentina MATEI[1], Mihaela BEGEA[1, 2], Valerica TUDOR, Mihai FRÎNCU[1], Răzvan Ionuț TEODORESCU[1]**

[1]University of Agronomic Sciences and Veterinary Medicine of Bucharest, 59 Marasti Blvd, District 1, Bucharest, Romania
[2]University Politehnica of Bucharest, 313 Splaiul Independentei, 060042 Bucharest, Romania

Corresponding authors email: ela_begea@yahoo.com

***Abstract***

*The present paper describes some studies regarding the potential of some conventional and non-conventional brewing and wine yeasts for their application in the craft beer process. The artisanal or the craft brewing industry has grown in the last decade. The representative microorganisms for the production of craft beers are Saccharomyces yeasts. Nevertheless, the non-Saccharomyces yeasts became important for the particular aromatic profile of craft beer, through the increased customer's experience and also for the potential health benefits such as the probiotic effect. The objectives of this paper were to screen the reported data regarding the general behavior of the microorganisms involved in craft beer production, such as the fermentative capacity to produced alcohol, flavors and the probiotic effects, including the general analytic assays. Starting from this research, studies will be opened for innovative pathways of using yeast strains isolated from vineyard and winery environment in artisanal beer production.*

***Key words****: Craft beer, yeast, Saccharomyces and non-Saccharomyces, brewing.*

## INTRODUCTION

Beer is an ancient beverage which is the most consumed alcoholic drink worldwide according to the World Health Organization (WHO) (Neffe-Skocińska et al., 2022). The history of beer is far back in times; Babylonians and Egyptians created a simple drink made from cereals let to germinate and after a spontaneously fermentation beer was brewed (Habschied et al., 2020).

Beer is a nutritious and refreshing carbonated drink produced from four types of ingredients according to the Purity Law adopted in Bavaria in 1516: water, barley, hops, and yeast. (Protz,R, 2004). Nowadays, a new trend is visible within the global beer market and apart from so-called industrial brewing industry, a new sector has been increased during recent years, that has been named the *craft beer* industry. For the forecast period (2022-2017) the compound annual growth rate (CAGR) for this business is estimated at 14.1% (Craft Beer Market, 2022)

In order to reach the higher expectations of customer in terms of taste, aroma, flavors, alcoholic, non-alcoholic, low alcoholic, low carbohydrates, gluten free, the craft sector grew progressively. This because a small brewery is more flexible and has the possibility to create innovative beers using different techniques, raw materials, and additional ingredients.

### Current trends on obtaining craft beer

Yeast has an important role in creating the distinctive aroma and flavor components which lead to an impressive variety of beer styles. Without the metabolism of yeast cells, the traditional production of alcoholic beverages would not be possible.

Beer production is divided in different styles, lager and ale being the most predominant: fermentation at the bottom on the fermenter (6-15$^{o}$C), performed by *Saccharomyces pastorianus* in lager beer production and top-fermented beer (temperatures between 16-24$^{o}$C) by the most studied yeast, *Saccharomyces cerevisiae*, producing ales. Ale beer is known for its fruity aromas, while lager beers are more neutral (Moura-Nunes et al., 2016).

Fermentation can be performed with *Saccharomyces* and *non-Saccharomyces* yeasts. Yeasts are largely responsible for the complexity and sensorial quality of fermented beverages. Choosing a pure yeast culture or a mix of strains is an important decision for achieving a final product which will have distinct characteristics appreciated by the consumers. The yeast is responsible to metabolize the sugar (maltose) in the wort into $CO_2$ and ethanol, but also is responsible for production through fermentation of by-products such as higher alcohols and esters, which have an important contribution to beer flavor.

It is known that *Saccharomyces* yeasts play an essential role in industrial brewing fermentation processes (Lengeler et al., 2020). Multiple studies have shown the major potential of *Saccharomyces* wild yeasts isolated from different foodstuffs to produce beer with new aromatic profile.

Postigo et al. (2021) conducted an evaluation of 141 *Saccharomyces* yeasts strains isolated from grapes and grape must. The main characteristics such as the fermentation behaviour, the production of volatile compounds, the melatonin production, and the antioxidant capacity were studied against the commercial strain *Saccharomyces cerevisiae*. The studies were carried from lab scale to industrial scale. In the end, *Saccharomyces*-G520 was indicated as a new potential yeast for the production of functional beer with healthier benefits, as this yeast can produce higher amount of melatonin and shows higher antioxidant capacity (Postigo et al., 2021).

The probiotic *Saccharomyces cerevisiae* var. *boulardii* (Scb) is a commercial strain of *Saccharomyces cerevisiae*, recently introduced in brewing industry. Mulero-Cerezo et al. (2019) used the probiotic *Saccharomyces cerevisiae* var. *boulardii* as a single yeast starter to produce craft beer with higher antioxidant activity and low alcohol content (Mulero-Cerezo et al., 2019). *Saccharomyces boulardii* is the most common and widely studied species with significant probiotic properties (Souza et al., 2022; Lazo-Velez et al., 2018). The use of *S. boulardii* in co-fermentation with *S. cerevisiae* to produce craft beers has been studied by Capece in 2018, who demonstrated that the probiotic yeast has the capacity to survive during the brewing process. The experiments performed by Capece refer to the integration of *S. cerevisiae* var. *boulardii* (S.b) in mixed cultures with *S. cerevisiae* strains for the production of beers which offer an increased content of polyphenols and higher antioxidant activity (Capace et al., 2018).

According to Datta et al. (2017), *S. boulardii* growth showed no significant difference in relation to the growth pattern presented by *S. cerevisiae* strains, although the probiotic strain showed higher tolerance to fermentative stress. *S. boulardii* had an antioxidant potential of 6 to 10 times higher than *S. cerevisiae*, with a total phenolic content 70 times higher and the total flavonoid content 20 times higher in the extracellular fraction. In addition, better results were obtained when *S. boulardii* and *Lactobacillus delbrueckii* acted together.

There is a continuous interest among research groups to achieve innovative fermentation characteristics from pure yeast cultures, that's why interspecific yeast hybrids are currently on focus for reinventing the lager yeast *Saccharomyces pastorianus* by crossing *Saccharomyces cerevisiae* with *Saccharomyces eubayanus* (Winans, 2022). Other authors, like Giannakou et al. (2021), created a novel non-GMO hybrid between *S. jurei* and *S. cerevisiae* ale yeasts to develop new starter strains with interesting flavours for the craft brewing.

Bruner et al. (2021) studied five distinct species of *Saccharomyces* from UC Davis selected from the Phaff Yeast Culture Collection, as well as an interspecies hybrid from the Fermentis company to produce beer on a pilot scale of 40 L. *S. kudriavzevii, S. mikitae, S. paradoxus, S. bayanus* and *S. uvarum* were inoculated in duplicate, with one fermenter in each pair receiving 10 g/L of dried hops during fermentation. The beers were analyzed for aroma, taste and mouthfeel, with the aim to assess the specific aroma of each yeast strain in terms of its brewing potential. All beers were spicy, probably due to the presence of phenols; the dry-hopping increased fruit notes at the same time perceived an increased bitterness and astringency.

**Assessment of the potential to use mixed yeast starter cultures in brewing**

The craft beer industry is interested in using new ingredients in order to develop new beer styles. According to this, the brewers' attention is focused to other, non-traditional method. Therefore, the *non-Saccharomyces* yeasts have been tested regarding their potential for aroma enhancement at the same time maintaining the specific *terroir* (Gamero et al., 2020).

***Non-Saccharomyces*** yeasts represent a group of microorganisms with genetic diversity, specific metabolic characteristics and high potential for use in different fermentation processes.. Either *non-Saccharomyces* or *Saccharomyces* yeasts share common pathways for the central metabolism of carbon; both groups metabolize glucose through glycolysis (Figure1, Steward, G., 2016).

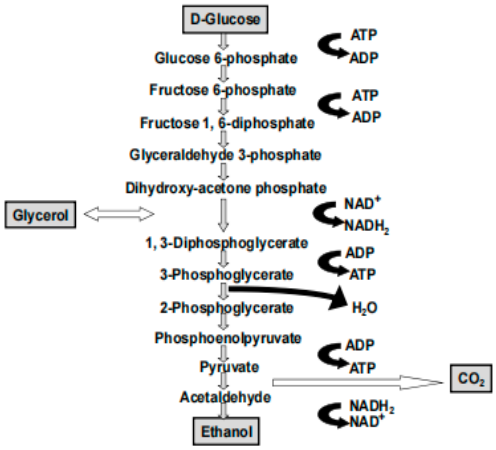


Figure 1. Obtaining of ethanol from glucose by the Embden-Meyerhof-Parnas (EMP) Pathway

Ethanol is the most important fermentation by-product, and from the technological point of view, the ethanol production capacity of yeasts is an important parameter that determines their use in fermentative processes (Steward, G., 2016).

The fermentative behavior of some non-Saccharomyces yeasts has been studied to find the most appropriate and suitable conditions for the strains to be used in the production of fermented beverages (Escalante, 2018). For a successful fermentation, a mixed culture with Saccharomyces spp. is required. It is necessary to search for a correct pitching ratio by testing different culture combinations at microscale, but also to scale-up the process to study the brewing process in real environment (Figure 2, Steward, G., 2016).

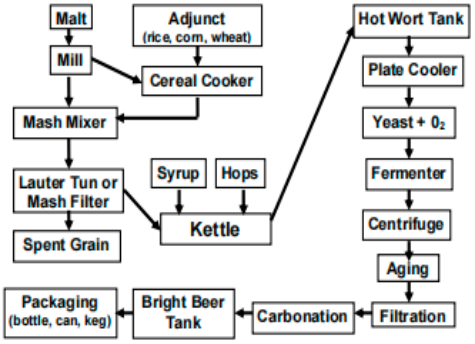


Figure 2. Beer brewing process

Non-*Saccharomyces* yeasts, such as *Dekkera animals, Naumovozyma dairenensis* and *Debaryomyces spp, Hanseniaspora, Torulaspora*, *Wickerhamomyces* have been reported with a high potential for use in beer fermentation because these yeasts can offer a diversified enzymatic apparatus and bioconversion abilities to reduce the energy value and lower the alcohol content in beer (Escalante, 2018).

*Cyberlindnera saturnus* TUM, a strain isolated from soil underneath an ash tree in Bavaria, was subject of a detailed studies made by Methner et al. (2022) about the influence of varying fermentation process to achieve different concentrations of flavors components in non-alcoholic beer. The trails were carried out using the surface methodology to optimize the beer with maximum content of fruitiness and minimal off-flavors and ethanol. The conclusion was that a low pitching rate combined with moderate fermentation temperature gave the optimal beer characteristics.

*Torulaspora delbrueckii* was explored in single strain fermentations for enhancing the aroma profile and producing a low alcohol beer. Some strains fermented all sugar, others did not succeed in this process. Important to mention is that all of them displayed an improved flavor profile

characterised by "fruit/citric" notes and "full-bodies" attribute, as well and enhanced clarity and persistent foam (Gamero et al., 2020).
Fermentation of barley-sorghum wort with *Saccharomyces cerevisiae, Torulaspora delbrueckii* and *Metschnikowia pulcherrima* yeast strains were studied (Einfalt, 2021). Diferences were noticed in terms of fermentation time and ability to ferment maltose. *S. cerevisiae* showed a high fermentative power, *T. delbrueckii* and *M. pulcherrima* have been found to have low maltose fermentation abilities and to offer significantly different sensory attributes to barley-sorghum beers.
Kveik yeast was subject of a recent study realized by Kawa-Rygielska et al. (2022). Kveik yeast is an unconventional Norwegian yeast used to make bread and produce New England India Pale Beer (NEIPA). The antioxidative activity of the final product described as the total phenolic compounds of malt, hop and melanoidins were studied. These compounds influence the key quality attributes of beer: clarity, color, taste, and storage stability. The antioxidative activity of worth and beers were analyzed and the higher antioxidative potential was reached by the strain Linda Kveik, an indicator which leads to interesting sensory features to be used in the growth of the segment of unconventional beer sector.

**Other current approaches in craft brewing**

Beer is a worldwide beverage, and the addition of different herbs is a well know procedure used especially by microbrewers. *Artemsia vulgaris, Juniperus communis, Melissa officinalis, Brasica nigra, Coraindrum sativum* are examples of plants and plant extracts used in brewing. Lemon juice, raspberry syrup and grapes are very popular through the women population who dislike the bitterness of the beer (Habschied et al., 2020).
*Ganoderma lucidum*, a medical mushroom has been studied by Leskosek-Cukalovic et al. (2010), in combination with beer by adding alcoholic extract. The final product gains in natural antioxidants and can have potential medical significance.
The continuous search for new functional beer brought to attention new raw materials such as buckwheat and amaranth for creating gluten free-beer (Dabija et al., 2022). The research performed to date shows the possibility of using of these pseudo cereals in different combinations to obtain new beer varieties.
Another trend is to use mixed microbial cultures such as yeast and lactic acid bacteria (LAB) to create sour beers. The last ones were considered contaminants in beer fermentation, nevertheless are also appreciated in production of sour beer and different wild beer styles, e.g. Lambics and American Coolship Ales (Piraine et al., 2021).
Craft beer is becoming more and more popular in Romania as new microbreweries, brewpubs and craft beer shops are opening (Figure 3, www.malt.ro).

Figure 3. Craft beer styles diversity

Innovation is mandatory in today's food industry and brewing industry is also in line with this trend. Customers are more interested in choosing themselves the food and drink products, and also to consider their nutritional and beneficial effects on health. Yeast starter culture is an important decision for craft brewing industry in designing functional beers with attractive sensorial characteristics.

## CONCLUSIONS

The "craft beer phenomenon" is one of the most fast growing trend in the beverages market worldwide, mainly because of the changes in consumer preferences and the interest for food and drinks with strong personality. One of the main characteristics of the sector that could be further explored is represented by the potential link with the local production of raw materials such as grains and hops, including the exploitation of

the local microflora that could be used either as pure culture or in mixed cultures with commercial yeasts for craft beer fermentation. For example, the use of different yeast strains *Saccharomyces* and non-*Sacharomyces* can contribute to obtain craft beers with diverse and attractive sensory characteristics, making possible the exploitation of the *terroir* concept in the brewing industry as well.

Starting from this research, studies will be opened for innovative pathways to use yeast strains isolated from vineyard and winery environment from the Pietroasa Viticulture and Winemaking Research and Development Station in the production of craft beers.

## ACKNOWLEDGEMENTS

This work was supported by the project 7PFE/2021 "Circular economy in USAMV farms - whole use of by-products resulting from fermentation processing", Program 1 - Development of the national research-development system, Subprogram 1.2 - Institutional performance - Institutional development projects - Projects to finance excellence in RDI.

## REFERENCES

Bruner, J., Marcus, A., & Fox, G. (2021). Brewing efficacy of non-conventional saccharomyces non-cerevisiae yeasts. *Beverages*, *7*(3), 68.

Capece, A., Romaniello, R., Pietrafesa, A., Siesto, G., Pietrafesa, R., Zambuto, M., & Romano, P. (2018). Use of saccharomyces cerevisiae var. boulardii in co-fermentations with S. cerevisiae for the production of craft beers with potential healthy value-added. *International Journal of Food Microbiology*, *284*, 22–30.

Coman, R. (2017). *13 craft beers you must try in Romania*. Culture Trip. Retrieved May 10, 2022, from /

*Craft beer market - growth, trends, COVID-19 impact, and forecasts (2022 - 2027)*. Craft Beer Market | 2022 - 27 | Industry Share, Size, Growth - Mordor Intelligence. (n.d.). Retrieved May 10, 2022

Dabija, A., Ciocan, M. E., Chetrariu, A., & Codină, G. G. (2022). Buckwheat and amaranth as raw materials for brewing, a review. *Plants*, *11*(6), 756.

Datta, S., Timson, D. J., & Annapure, U. S. (2017). Antioxidant properties and global metabolite screening of the probiotic yeast*saccharomyces cerevisiae*var.*boulardii*. *Journal of the Science of Food and Agriculture*, *97*(9), 3039–3049.

Escalante, W. D. E. (2019). Perspectives and uses of Non-Saccharomyces yeasts in fermented beverages. *Frontiers and New Trends in the Science of Fermented Food and Beverages, 107*.

Einfalt, D. (2020). Barley-sorghum craft beer production with saccharomyces cerevisiae, Torulaspora delbrueckii and Metschnikowia pulcherrima yeast strains. *European Food Research and Technology*, *247*(2), 385–393.

Gamero, A., Dijkstra, A., Smit, B., & de Jong, C. (2020). Aromatic potential of diverse non-conventional yeast species for winemaking and brewing. *Fermentation*, *6*(2), 50.

Giannakou, K., Visinoni, F., Zhang, P., Nathoo, N., Jones, P., Cotterrell, M., Vrhovsek, U., & Delneri, D. (2021). Biotechnological exploitation of Saccharomyces Jurei and its hybrids in craft beer fermentation uncovers new aroma combinations. *Food Microbiology*, *100*, 103838.

Habschied, K., Živković, A., Krstanović, V., & Mastanjević, K. (2020). Functional beer – a review on possibilities. *Beverages*, *6*(3), 51.

Kawa-Rygielska, J., Adamenko, K., Pietrzak, W., Paszkot, J., Głowacki, A., & Gasiński, A. (2022). Characteristics of new england india pale ale beer produced with the use of Norwegian kveik yeast. *Molecules*, *27*(7), 2291.

Lazo-Vélez, M. A., Serna-Saldívar, S. O., Rosales-Medina, M. F., Tinoco-Alvear, M., & Briones-García, M. (2018). Application of saccharomyces cerevisiae var. Boulardii in Food Processing: A Review. *Journal of Applied Microbiology*, *125*(4), 943–951.

Leskosek-Cukalovic, I., Despotovic, S., Lakic, N., Niksic, M., Nedovic, V., & Tesevic, V. (2010). Ganoderma lucidum – medical mushroom as a raw material for beer with enhanced functional properties. *Food Research International*, *43*(9), 2262–2269.

Methner, Y., Dancker, P., Maier, R., Latorre, M., Hutzler, M., Zarnkow, M., Steinhaus, M., Libkind, D., Frank, S., & Jacob, F. (2022). Influence of varying fermentation parameters of the yeast strain Cyberlindnera Saturnus on the concentrations of selected flavor components in non-alcoholic beer focusing on (e)-β-damascenone. *Foods*, *11*(7), 1038.

Moura-Nunes, N., Brito, T. C., Fonseca, N. D., de Aguiar, P. F., Monteiro, M., Perrone, D., & Torres, A. G. (2016). Phenolic compounds of Brazilian beers from different types and styles and application of chemometrics for modeling antioxidant capacity. *Food Chemistry*, *199*, 105–113.

Mulero-Cerezo, J., Briz-Redón, Á., & Serrano-Aroca, Á. (2019). Saccharomyces cerevisiae var. Boulardii: Valuable probiotic starter for craft beer production. *Applied Sciences*, *9*(16), 3250.

Neffe-Skocińska, K., Kruk, M., Ścibisz, I., & Zielińska, D. (2022). The novel strain of Gluconobacter oxydans H32 isolated from Kombucha as a proposition of a starter culture for sour ale craft beer production. *Applied Sciences*, *12*(6), 3047.

Piraine, R. E., Leite, F. P., & Bochman, M. L. (2021). Mixed-culture metagenomics of the microbes making sour beer. *Fermentation*, *7*(3), 174.

Postigo, V., García, M., Cabellos, J. M., & Arroyo, T. (2021). Wine saccharomyces yeasts for beer fermentation. *Fermentation*, *7*(4), 290.

Protz, Roger.*The Complete Guide to World Beer*, 2004

Souza, H. F., Carosia, M. F., Pinheiro, C., Carvalho, M. V., Oliveira, C. A., & Kamimura, E. S. (2022). On probiotic yeasts in food development: Saccharomyces boulardii, a trend. *Food Science and Technology*, *42*.

Stewart, G. (2016). Saccharomyces species in the production of beer. *Beverages*, *2*(4), 34.

Winans, M. J. (2022). Yeast hybrids in brewing. *Fermentation*, *8*(2), 87.

www.malt.ro