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FLOCCULATION PROCESS OF YEAST BREWERS®

Proces flokulacji drożdży piwowarskich®

Yeast flocculation is a reversible, asexual and calcium-dependent process by which cells stick to form flocs of thousands of cells. After the formation of a larger cell conglomerate, there is a massive separation from the medium by sedimentation of the bottom fermenting yeast. The ability to flocculate yeast cells is of great importance to the brewing industry as it effectively separates the yeast cell from the green beer at the end of fermentation. Therefore, a strong and complete flocculation property of the brewer's yeast is desired. However, yeast cells should not flocculate too quickly until the wort is completely fermented as premature flocculation slows down fermentation and can also lead to inappropriate volatile components. Therefore, ideal brewer's yeast should flocculate strongly towards the end of fermentation. This flocculation should also be constant in subsequent fermentations. ***The phenomenon of flocculation is a complex process influenced by biochemical, genetic and physical mechanisms.***

Key words: yeast, flocculation, fermentation, yeast harvest.

Flokulacja drożdży jest odwracalnym, bezpłciowym i zależnym od zawartości wapnia procesem, w którym komórki przylegają, tworząc kłaczki składające się z tysięcy komórek. Po utworzeniu większego konglomeratu komórek, następuje masowe oddzielenie od podłoża przez sedymentację drożdży dolnej fermentacji. Umiejętność flokulacji komórek drożdży ma duże znaczenie dla branży piwowarskiej, ponieważ zapewnia w skuteczny sposób oddzielanie komórkę drożdży z zielonego piwa pod koniec fermentacji – dlatego pożądana jest silna i pełna właściwość flokulacji drożdży piwowarskich. Komórki drożdży nie powinny jednak zbyt szybko flokulować, zanim brzeczka nie będzie całkowicie odfermentowana, gdyż przedwczesna flokulacja powoduje spowolnienie fermentacji, a także może prowadzić do uzyskania niewłaściwych zawartości lotnych komponentów. Idealne drożdże piwne powinny się charakteryzować silną flokulacją pod koniec fermentacji i powinna ona być również stała w kolejnych fermentacjach. ***Zjawisko flokulacji jest procesem złożonym, na które mają wpływ mechanizmy biochemiczne, genetyczne i fizyczne****.*

Słowa kluczowe: drożdże, flokulacja, fermentacja, odbiory drożdży.

INTRODUCTION

The main aim of brewer's wort fermentations are to consistently metabolise wort into ethanol, carbon dioxide, glycerol, and other fermentation products, many of which contribute to beer flavour in order to produce it with satisfactory quality, drinkability, and stability of beer. Another very important fermentation objective is to produce yeast crops that can be harvested, stored, and subsequently re-pitched into a later brew. The most important features of yeast should be:

- yeast tolerance to stress factors such as: heat, osmotic pressure, temperature, ethanol, mechanical tension,
- wort gravity,
- wort dissolved oxygen concentration at pitching and during fermentation,
- flocculation, adhesion, sedimentation, and harvesting characteristics,
- culture characteristics during storage between fermentations and its subsequent re-pitching into wort [8].

In the article, will be underline very important issue of flocculation of brewery yeast. Flocculation is a complex phenomenon that is affected by genetic, physiological, and environmental factors. Within the two brewer's yeast species - Saccharomyces cerevisiae (ale) and Saccharomyces pastorianus (lager) there are both flocculent and non-flocculent strains. Flocculation is an import ant characteristic in an environment with limited nutrients because the death and autolysis of the cells inside flocs can provide further nutrients to cells in the surrounding environment. Flocculation may enhance the survival of yeast cells during adverse conditions.

Genetic studies on yeast flocculation began over 70 years ago. The first publication on this subject was by Pomper and Burkholder, who reported crossing a haploid culture that possessed a "dispersed" character (non-flocculent) with a haploid culture that possessed a non-dispersed charakter (flocculent). The "dispersed" character was reported to be dominant over the non-dispersed character. In the early 1950s, Gilliland (working in the Guinness laboratory in Dublin) and Thorne (working in the Alfred Jorgensen laboratory in

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Copenhagen) independently carried out extensive studies on the genetics of yeast flocculation. Gilliland studied two non-brewing strains of S. cerevisiae which differed only in their flocculation properties. It was proposed that a single gene was responsible for the flocculent phenotype. Thorne confirmed Gilliland's studies and demonstrated that yeast flocculence was an inherited characteristic and that the flocculence phenotype was dominant over the non-flocculent characteristic. These studies were also confirmed by Guo et al. [8].

FLOCCULATION OF YEAST

Flocculation of yeast cells is usually observed at the end of beer fermentation and is of great importance in brewery. It happened, when wort is almost attenuated and process of fermentation is finished (Fig.1).

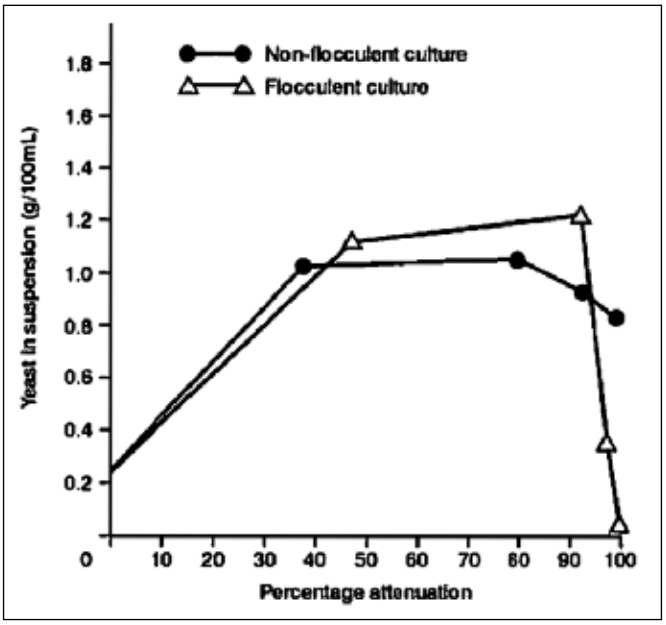

Fig. 1. Static fermentation flocculation characteristics.
Rys. 1. Charakterystyka flokulacji po zakończeniu fermentacji.

Source: Lekkas i in. 2007 [2]
Źródło: Lekkas i in. 2007 [2]

Its mechanism is not yet completely understood and is still the subject of much controversy. It is a very complex process which depends on numerous factors: the yeast strain especially genetics, the composition of the wort, and the culture conditions [4].

It has been shown that some highly flocculent cultures lost their flocculation characteristic during the early stages of growth, in the presence of nutrients, and recovered it towards the end of the exponential growth phase coinciding with nutrient depletion [8].

The issues for, and advantages of, yeast flocculation (particularly, but not only, during the brewing process) have been extensively studied and deliberated. Flocculation may enhance the survival of yeast cells during adverse (starvation and autolysy) conditions. Flocculation is an important characteristic in an environment with limited nutrients because the death and autolysis of the cells inside flocs can provide further nutrients to cells in the surrounding environment to beer. It has been shown that some highly flocculent cultures lost their flocculation characteristic during the early stages of growth, in the presence of nutrients, and recovered it towards the end of the exponential growth phase coinciding with nutrient depletion [1, 3, 6].

As with many aspects of brewer's yeast metabolism, yeast flocculation is rather primarily strain-dependent. During the later stages of fermentation, the following conditions are favourable for culture sedimentation:

- the carbon dioxide production rate is slow,
- wort attenuation is approaching completion—most of the fermentable sugars in wort have been consumed by the yeast culture, including glucose, fructose, sucrose, maltose, and finally maltotriose,
- flocculation ability is high but not too much,
- yeast concentration in suspension is maximal.

There are several factors that influence the rate of floc sedimentation out of the wort:

- The way that yeast cells are packed into flocs,
- the floc size, shape, and concentration (density),
- nurture factors that include wort properties and encompasses: concentration (gravity), viscosity, density, and natural convection in fermentor,
- higher gravity worts, following fermentation, results in "green" (immature) beers with a higher viscosity and density. Both these factors will retard yeast sedimentation and lead to increased osmotic pressure and ethanol prior to dilution to the fermented wort's sales gravity and alcohol concentration. The floc size often decreases as settling proceeds because the concentration of yeast cells in the suspension continues to decrease and smaller flocs form which will slowly settle out of the suspension [7, 8].

A few factors can affect yeast flocculation. They are very important in area of yeast management, especially for yeast flocculation (Fig. 2).

The first factor is the genetic background of a strain. Flocculin proteins are encoded by members of the FLO group of genes. The genetic background (nature effects) with regard to FLO genes varies greatly amongst various types of brewer's yeast (ale or lager) and other strains. Strains contain a variety of FLO gene combinations, resulting in a spectrum of flocculation characteristics.

Second, flocculation is caused by the physiological environment. This includes the pH, availability of appropriate metal ions, and nutrients during the growth phase. The pH will influence the cell surface charge which will have an effect on the flocculation phenotype because the environment is sensed by cells leading to the expression of FLO, their translation to Flo proteins, and their location into the cell wall, which is influenced by a number of environmental factors. Changes in pH may also modify the ionisation of functional groups in flocculin proteins, which will modify their conformation.

Flocculation of yeast cells involves lectin-like proteins – so-called flocculins – that stick out of the cell walls of flocculent cells and selectively bind mannose residues present in the cell walls of adjacent yeast cells (Fig. 3). Calcium ions in the medium are needed in order to activate the flocculins [11].

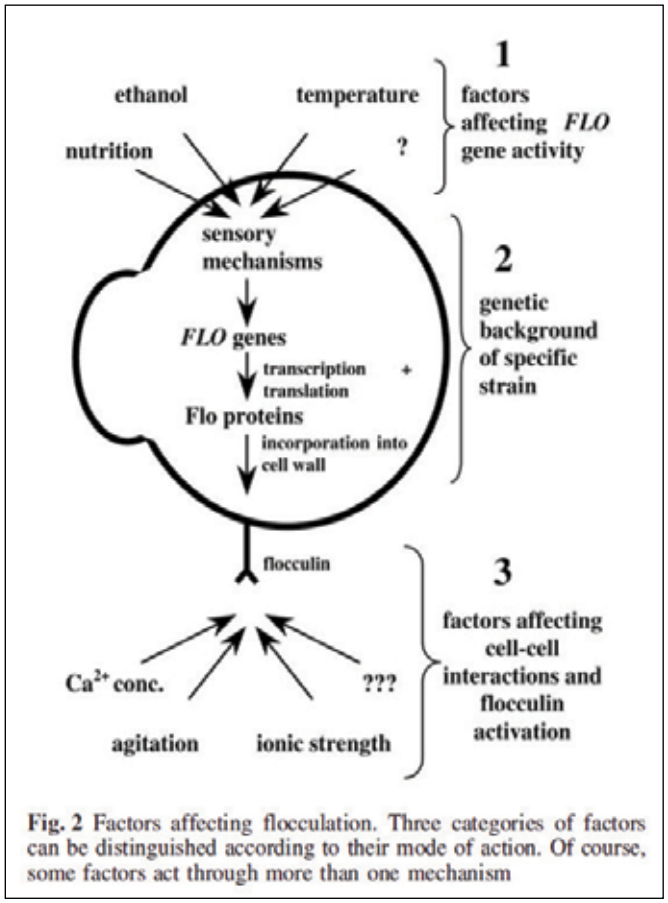

Fig. 2 Factors affecting flocculation. Three categories of factors can be distinguished according to their mode of action. Of course, some factors act through more than one mechanism

Fig. 2. Factors affecting flocculation.
Rys. 2. Czynniki wpływające na flokulację.
Source: Verstrepen i in. 2003 [11]
Źródło: Verstrepen i in. 2003 [11]

Finally thirdly, the physical environment affects flocculation. The hydrodynamic (relevant to liquids in motion) conditions must be favourable and promote collision rates between cells. However, natural convection in fermentor must not be sufficiently violent to cause flocs to disperse. In addition, there must be sufficient cells in suspension in order to result in cell collisions to form floks [5, 8, 9, 10].

Brewer's yeast flocculation is an off-cost process which does not require significant energy input. However, cooling at the end of fermentation does facilitate yeast separation and this normally requires some energy. It is considered that flocculation increases process efficiency and reduces the energy consumption [8].

CONCLUSION

The yeast flocculation process is a very important phenomenon in brewing. After fermentation, the lager yeast should naturally sediment to the bottom of the fermentation tank. The process should be timely - neither too early nor too late. Premature flocculation interrupts fermentation, while too long sedimentation deteriorates yeast harvest from the fermenter, which results in a low concentration of collected yeast, and those cells that remain in the beer contribute to the unfavorable autolysis phenomenon. The phenomenon of yeast flocculation is influenced by three factors, of which genetics are the most important. Only in the case of the content of metal ions, and in particular the amount of calcium, can its content in the fermenting wort be adjusted in any way.

PODSUMOWANIE

Proces flokulacji drożdży jest bardzo istotnym zjawiskiem w piwowarstwie. Drożdże dolnej fermentacji po zakończonej fermentacji w sposób naturalny powinny sedymentować na dno zbiornika fermentacyjnego. Proces ten powinien przebiegać we właściwym momencie – ani za wcześnie ani za późno. Przedwczesne kłaczkowanie powoduje przerwanie fermentacji, natomiast zbyt długie sedymentowanie wpływa na pogorszenie odbiorów drożdży z fermentora, co powoduje niską koncentrację zebranych drożdży a te komórki drożdży które pozostają w piwie, przyczyniają się do niekorzystnego zjawiska autolizy. Na zjawisko flokulacji drożdży mają wpływ trzy uwarunkowania, z których najbardziej istotnym są uwarunkowania genetyczne. Jedynie w przypadku zawartości jonów metali, a zwłaszcza wapnia, można w dowolny sposób regulować jego zawartość w fermentującej brzeczce.

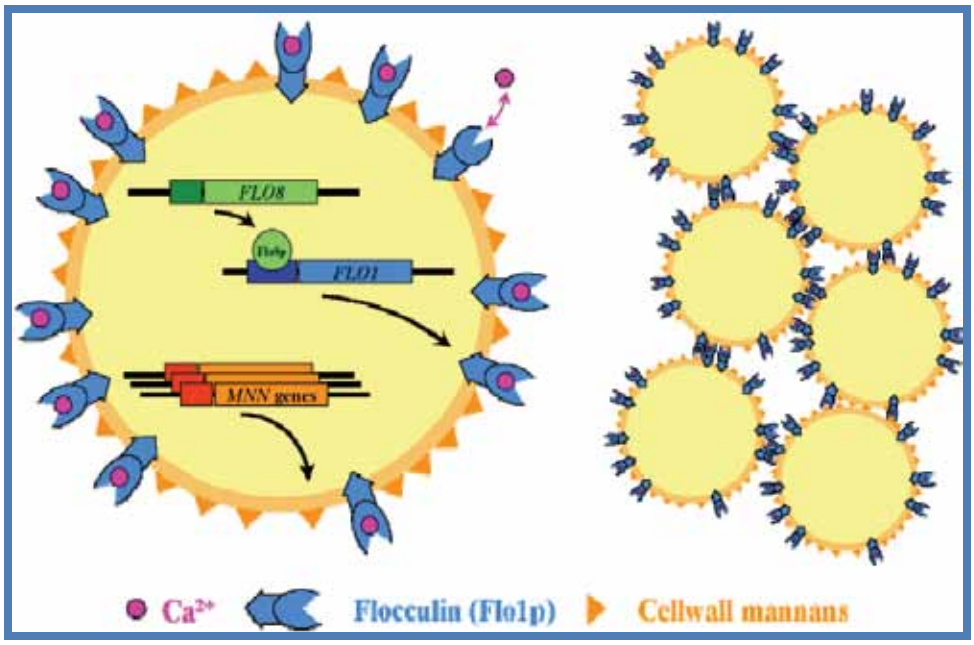

Fig. 3. Simplified lectin-theory of yeast flocculation.
Rys. 3. Uproszczona teoria wpływu lektyn na flokulację drożdży.
Source: Voetz M. 2008 [12]
Źródło: Voetz M. 2008 [12]

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