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Effect of hops addition to the fermentation tank on beer fermentation

BREWING SUMMIT SAN DIEGO 2018

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Introduction

We previously reported that the addition of hops to a fermentation tank produces beer with a strong hoppy aroma (linalool) and minimizes unpleasant flavors derived from hops (myrcene). The fermentation technology used to exploit this phenomenon, known as the **'dip hop'** method, was implemented by the Kirin group **in 2012** (EBC2013). **In this study, we investigated the effects of the 'dip hop' method on fermentation and the production of 2-mercapto-3-methyl-1-butanol (2M3MB, onion-like off-flavor).**

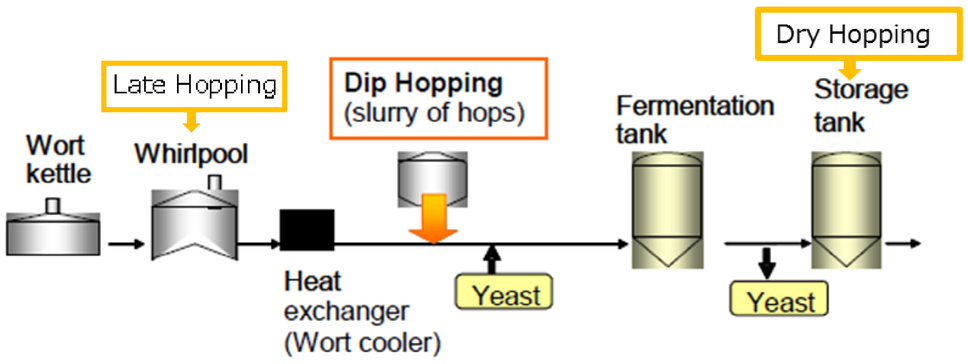


Fig. 1 Dip-hopping procedure

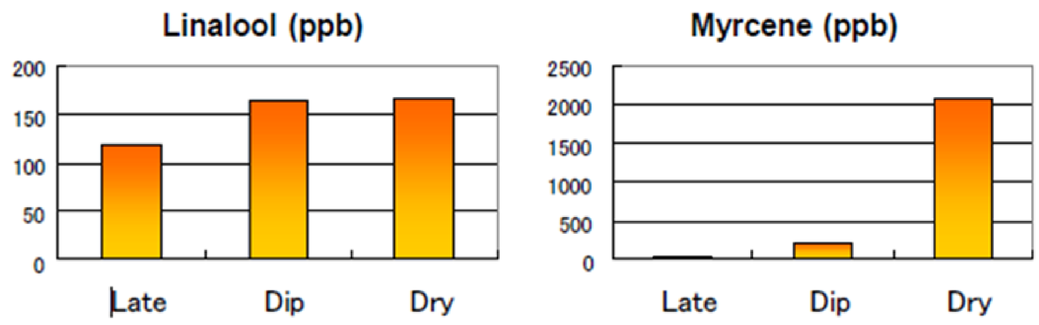


Fig. 2 Linalool and myrcene content of late, dip, and dry hopped beer

Materials and methods

The condition of the wort used for fermentation tests was assessed in three trials: Trial 1: no 'dip hopping', Trial 2: 'dip hopping' (3 g/L), Trial 3: activated carbon addition (3 g/L); activated carbon was added to the wort to examine the effect of fermentation on the solids content itself.
Fermentation conditions involved pitching with 10^7 cells/ml at 12°C in a 200 L pilot plant.

Results and Discussion

Fermentation characteristics obtained using the 'dip hop' method

Result 1: Accelerated fermentation

Fermentation was conducted with and without dip hopping. Dip hopping increased the sugar consumption rate and the number of yeast cells, compared with no dip hopping. Similar results were obtained by adding activated charcoal to the wort.

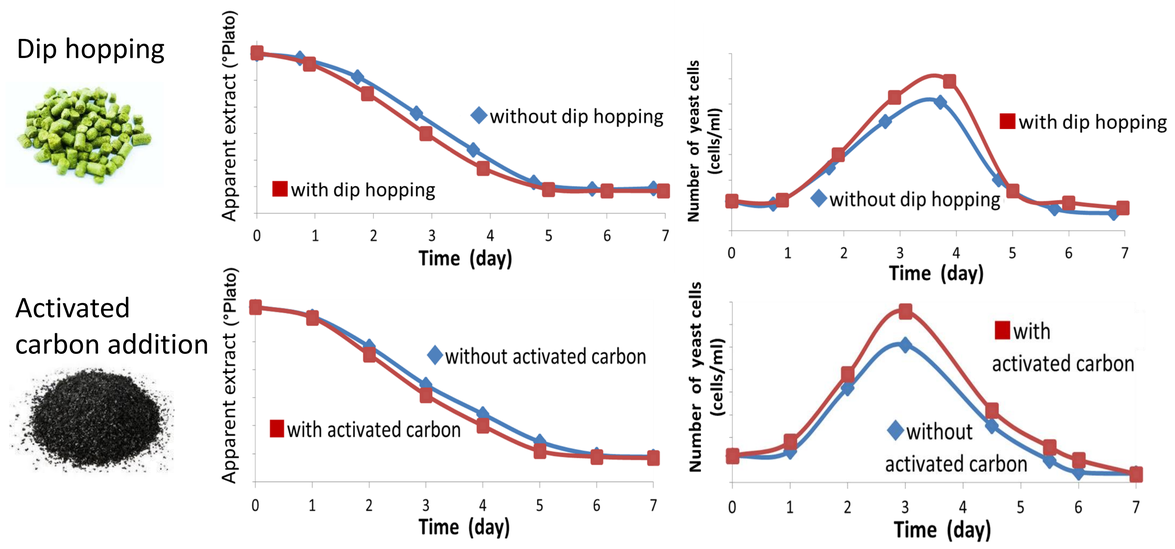


Fig. 3 Changes in apparent extract and number of yeast cells during fermentation

Which factors promote fermentation?

(1) Hops enzymes do **NOT** contribute markedly to fermentation .

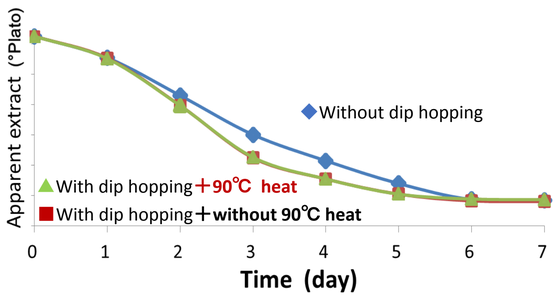


Fig. 4 Fermentation profiles

The enzymes in hops have low glycolytic activities; the fermentation-promoting effect of the dip hop method was observed even when hops were boiled, implying that the contribution of hop-associated enzyme activity was considered to be small.

(2) The reduction in DCO_2 by dip hopping markedly affects fermentation.

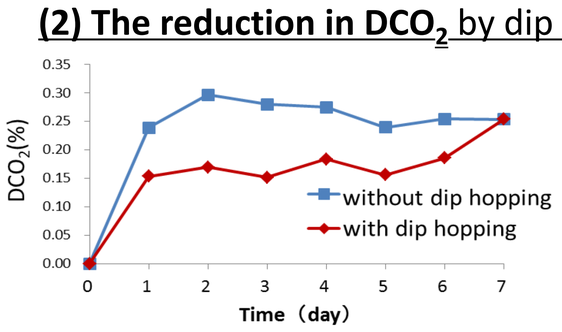


Fig. 5 Change in the concentration of DCO_2

Supersaturated DCO_2 is known to inhibit yeast growth, [1] and dip hopping reduces the concentration of DCO_2. The hydrophobic group of added solids promotes bubbling of DCO_2 and the DCO_2 decreases.

These results suggest that the fermentation-promoting effect of the dip hop method is attributed to the hops particles themselves.

Result 2: Reduction of 2M3MB; an onion-like off-flavor in beer.

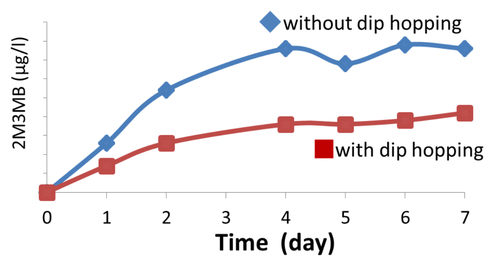


Fig. 6 2M3MB levels during fermentation

Decreased production of 2M3MB derived from fermentation was also observed.

What causes the lower production of 2M3MB with dip hopping?

The formation of H_2S is key to the formation of 2M3MB in beer.

iso-α-acid
⇩
2,3-epoxy-3-methyl-butanal (EMB) [2]
⇩ ⇦ ***hydrogen sulfide (H_2S)***
⇩
2M3MB

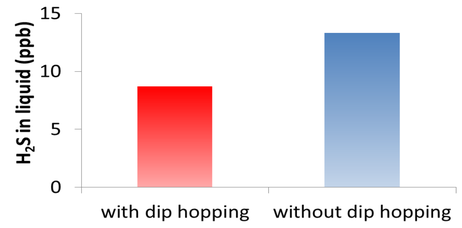
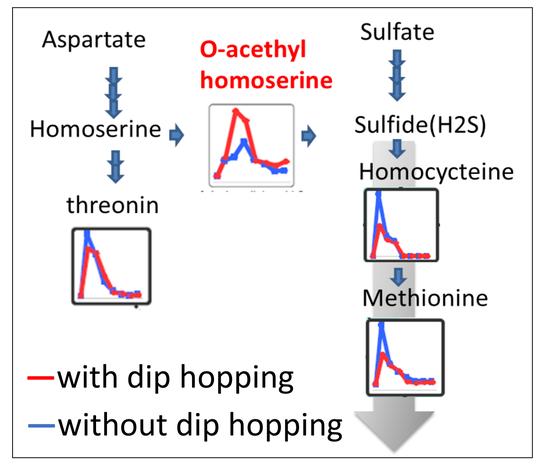


Fig. 7 H_2S content of young beer

What factors cause a reduction in H_2S ?

Hypothesis (1):
H_2S decreases due purging by CO_2.

Hypothesis (2):
Metabolome analysis revealed that in sulfur metabolism, intracellular threonine is more easily consumed by dip hopping, resulting in increased levels of O-acetyl homoserine (OAH).
By increasing OAH, the pathway from H_2S to homocysteine is activated and H_2S production decreases.[3]



Analyzed by Human Metabolome Technologies Co., Ltd.

Fig. 8 Yeast sulfur metabolism

Conclusion

Brewing beer using the dip hop method imparts a pleasant hoppy aroma to the beer by suppressing off-flavors derived from fermentation. The method emphasizes the aroma of beer without increasing the unpleasant flavor of myrcene.

By using the **dip hop** method, we developed the beer **"GRAND KIRIN"**

References:
1) Johnes RP, Greenfield, PF: Enzyme Microb. Technol., 4, 210, (1982).
2) Noba S et al., Asahi Breweries, Ltd.: Elucidation of the formation mechanism of 2M3MB in beer (EBC 2017).
3) Yoshida S et al., Kirin co, Ltd.: Development of bottom-fermenting Saccharomyces strains that produce high SO_2 levels using integrated metabolome and transcriptome analysis. Appl Environ Microbiol.;74(9):2787-96(2008).