PROCESS CONTROL FOR HIGHER ALCOHOLS IN BEER: YEAST & BILL OF MATERIALS

Yeast Genetics

TFL resistant mutants (esp. isoamyl alcohol) (+)

Carlsberg: SSY1 gene produces the "sensing" protein on the cell surface involved in triggering the production of amino acid permeases.

Catabolic pathway produces all higher alcohols *except* for **n-propanol**.....**only anabolic** generates it!

Strain (very important!), levels of individual alcohols can vary by 500%!).

4-vinyl guaiacol, a phenol (PDF$^+$, yeast, decarboxylate malt ferulic acid, creating 4-VG and the "clove" flavor).

Catabolic pathway.....early in fermentation **Anabolic Pathway**.....later in fermentation.

Suntory: if create lager yeast with the *BAP2* gene (for amino acid permease) being constitutive, saw beer with higher levels of isoamyl alcohol from leucine...did not see same effect for isobutanol (from valine) or active amyl alcohol (from isoleucine).

Deletion of BAT2 gene in ILV pathway results in the production of very high levels of higher alcohols and VDK.

Yeast Physiology

Yeast crop (+)

Higher alcohols are the by-products of both synthesis and catabolism of amino acids, especially isoleucine, leucine and valine.

Valine...isobutanol.

Leucine....isoamyl alcohol.

Anything promoting yeast growth (+). CB: "Basically factors promoting yeast growth favor the production of higher alcohols, but suppress the development of esters."

Isoleucine....active amyl alcohol.

"Higher alcohols, especially phenylethanol, produced at same rate as CHOs and a.a.'s are assimilated".

High level of **Group A** amino acids as suppresses synthesis of isoleucine/valine/leucine (-)

Maltose grown cells yield higher levels than glucose or fructose grown cells.

Maximum production when concentration of assimilable N_2 is growth limiting (+) (via Biosynthetic Pathway).

Quantity of FAN consumed during fermentation (+)

Levels directly related to amount of yeast growth.

Reduction of malt aldehydes derived from LOX action (e.g. hexanal, hexanol) or Maillard/Strecker reactions (e.g. 3-methyl butanal, 3-methyl butanol).

Anabolic pathway of formation (+/-)

Catabolic pathwayof formation (+/-)

Lie and Haukeli now believe higher alcohols mostly made when yeast growth in numbers stops, and switches to increasing biomass by glycogen accumulation…"formation is closely correlated to ethanol formation, and not to yeast growth".

Effect on Levels of Higher Alcohols

Suntory: if **add valine/isoleucine/leucine to wort** see increase in [ ] of their higher alcohols in beer via assimilation of these amino acids.

Kirin: Saw higher levels of higher alcohols in adjunct vs. all malt brewing... 250 vs. 125 ppm, respectively.

Trub (+)

Zinc (+)

Production of the **aromatic alcohols**, phenylethanol (pleasant fragrance), tyrosol (bitter, chemical) and tryptophol (solvent, almond) increases at higher temperatures or very low **FAN worts**….latter two are negative flavors.

Barley variety, growing region (+/-)

High [MALT] decreases phenylethanol.

High C:N ratio (-)

High [FAN] decreases phenylethanol.

Relative concentrations of aminoacids (Ehrlich).

Isoleucine, valine, leucine (+)

Higher wort OG's yield higher [yeast produced **glycerol**], especially lager vs. ale yeast.

Overmodified malts increase [isoamyl alcohol].

Higher FANs decrease anabolic amino acid synthesis, while cataboloic mechanism for higher alcohols increases.....net result of this shift is that as [FAN] increases - with everything else constant - see gradual decline in [higher alcohols] with increasing wort FAN.

Total [higher alcohols] produced **linearly related to extent of yeast growth**. Anything promoting growth (e.g. D.O., FAN, etc...) increases their production.

Undermodified malts increases [isopropanol].

Very high or very low assimilable N_2 (+) compared to standard FAN levels.

Suntory: used **Gene Expression Microarray technology** to study gene expression in all-malt and 25% adjunct brews...distinct gene expression profiles, fermentation profile and formation of esters & alcohols differed significantly.....using it to study gene expression/formation of flavor volatiles....**fusels much higher in adjunct brew**, especially isobutanol and isoamyl alcohol....believed due to elevated anabolic pathway.

As percent maltose in extract increases, [higher alcohols] decreases.

HGB (+, disproportional for n-propanol, n-butanol) (isoamyl alcohol, isobutanol made in proportion to increase in ethanol & OG).

Bill of Materials

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ASBC No. XIVa
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