NEER™ is frozen Pichia kluyveri for direct inoculation that produces <0.5% alcohol only from monosaccharides

Fermentation characteristics

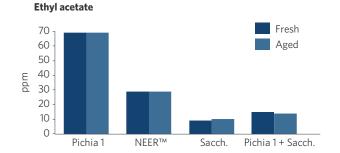
- > Enhanced fruit flavors: high production of esters and thiols without diacetyl
- > Medium production of polysaccharides
- > Low ethanol production
- > Low pH drop

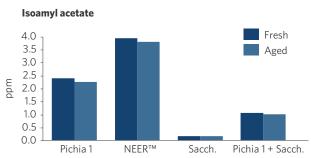
Main benefits of NEER™

- Flavorful compounds from the specially isolated and selected Pichia kluyveri strain
- No propagation equipment or slant procedures required direct inoculation
- Frozen concept means that NEER™ is always ready when you need it

Application







NEER™ Pichia kluyveri for production of non-alcoholic beer

Introduction

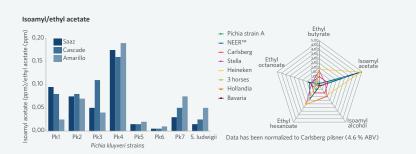
Due to increased consumer awareness and a desire to live healthier, the global beer market is experiencing a large increase in sales of low- (< 3.5 % ABV), and non-alcoholic beers (< 0.5 % ABV), especially during the last 5 years. However, most non-alcoholic beers suffer from inferior organoleptic properties due to the 'side-effects' of the physical removal of ethanol or the application of cold-contact with regular brewers' yeasts. We here present a TRUE brewing solution, NEER™, for the production of full bodied, flavorful, non-alcoholic beers, based on the application of our patented *Pichia kluyveri* strain. NEER™ offers a low CAPEX and OPEX alternative compared to the newer technical methods for production of non-alcoholic beers, e.g. membrane removal of ethanol. Furthermore, NEER™ is sold in a direct pitch format, hereby circumventing the need for propagation. NEER™ is, to our knowledge, the only yeast for production of non-alcoholic beers sold in this convenient format.

P. kluyveri isolation and characterization

A large array of *P. kluyveri* strains were screened based on the following criteria:

- Maltose negative strains
- > No off-flavor production
- > No/very low alcohol production
- Tolerate hop compounds
- > High level of esters
- > Low to moderate oxygen demands

Subsequently, the 7 best candidates were tested for their ester production capabilities in combination with 3 different hop varieties. From this experiment (bar plot) the best *P. kluyveri* strain (Pk4 -> NEER™) was selected. When this strain was tested in a standard pilsner wort, it produced a beer with a positive flavor profile (spider web plot).



From freezer to fermentor in 1 hour









NEER™ concept

NEER™ is produced using Chr. Hansen's proprietary production method which ensures high product performance, consistency and low risk of contamination. The NEER™ product is delivered on dry-ice as a 1 kg frozen block that after thawing can be transferred to the fermentation vessel using the sterile connection tube included in the package. NEER™ is highly concentrated and 1 bag (1 kg) can be used to inoculate 500 hL of wort and normally finishes fermentation within 3-5 days. Minimal maturation of the beer is required.

Brewing process comparison

Compared to a regular brewing process, NEER™ can substitute brewers' yeast without having to make significant changes to the process. NEER™ only assimilates monosaccharides and as a result some adjustments have to be made:

- > The fermentation tank needs to be mixed as lower level of produced CO₂ limits fluid motion in the tank.
- The mashing regime and malt utilization needs to be tuned so the resulting wort has ~10 g/L monosaccharides content. Higher monosaccharide levels can be used, but then the fermentation needs to be stopped by cooling to avoid overshooting 0.5 % ABV or a subsequent dilution step with de-aerated water implemented. The extend of the respiratory phase also influences the final alcohol level.
- angle Due to the residual oligosaccharides in the final beer, high QC standards or bottle pasteurization have to be implemented.

NEER™ has been demonstrated to be applicable for producing non-alcohol version of almost any beer style.

Pre-fermentation Fermentation Post-fermentation Low/high gravity Maturation, filtering, Trad. full alc. Propagation bottling fermentation Mashing, lautering, Direct inoculation of Centrifugation or NFFR™ hee boiling, P. kluyveri filtration, bottling whirlpooling Interrupted or halted Trad. low alc. Physical removal of Propagation fermentation

"The Pichia trial we did at our end produced some fantastic ester aromas and really helped to kill off the awful wort character of typical N/A beer"

- Verified customer quote

Simon Carlsen†, David Spector ‡ and Sofie M.G. Saerens† † Department of Wine and Fermented Beverages, Chr. Hansen A/S. Denmark.

‡ Cultures and Enzymes, Chr. Hansen A/S, USA

For more information, please visit www.chr-hansen.com



The information contained herein is presented in good faith and is, to the best of our knowledge and belief, true and reliable. It is offered solely for your consideration, testing and evaluation, and is subject to change without prior and further notice unless otherwise required by law or agreed upon in writing. There is no warranty being extended as to its accuracy, completeness, currentness, noninfringement, merchantability or fitness for a particular purpose. To the best of our knowledge and belief, the product(s) mentioned herein do(es) not infringe the intellectual property rights of any third party. The product(s) may be covered by pending or issued patents, registered or unregistered trademarks, or similar intellectual property rights. All rights reserved.