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KOMBUCHA:- FERMENTED TEA AND BACTERIAL CELLULOSE

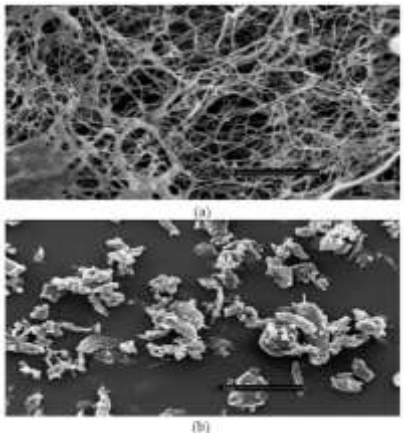
Aim – *To investigate properties affecting the conversion of sucrose to organic acids and bacterial cellulose in the kombucha tea fermentation process.*

Kombucha – fermented tea

Kombucha is a traditional drink made by fermenting tea sweetened with sucrose sugar. The fermentation is performed by a symbiotic culture of bacteria and yeast (SCoBY) in which the yeasts (typically *Saccharomyces cerevisiae* and others) convert the sucrose to glucose and fructose, and further to ethanol and carbon dioxide. In turn the bacteria (particularly *Komagataeibacter xylinus*) convert the ethanol to acetic acid and glucose to gluconic acid. In addition the *K. xylinus* produces a cellulose network at the surface that restricts the access of oxygen and unwanted contaminants. The tea is popular as a refreshing drink with supposed health benefits.

Bacterial cellulose

The *Komagataeibacter xylinus* bacteria that is present in Kombucha is reported to have amongst the highest recorded rates of production of bacterial cellulose. The structure of the network is also of interest with respect to production of nanoscaled biocomposites.



SEM micrographs a) bacterial cellulose from kombucha (scale bar 5 µm) and b) commercial microcrystalline cellulose (scale bar 20 µm), [1]

Propagation of kombucha culture

The Kombucha tea culture has survived over two thousand years by batch cyclic production and transfer from household to household. For each new batch about 10-15% of the previous batch is used as an inoculant. It can be expected that the culture evolves by incorporating local microorganisms. Regional differences in the consortium have been reported [2], and it has also been claimed that the conversion efficiency of household cultures is poorer than commercial ('pharmacy') cultures, [3]. In addition a widely cited study [4] claimed that fructose is not utilised in the fermentation. To investigate these claims a lab trial was performed at NORUT in July 2016 by a summer student, Rajina Yogarajah.

Fermentation trial

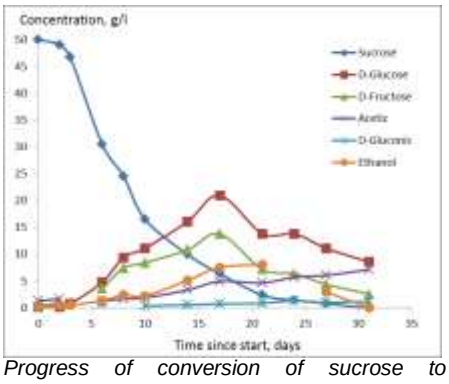
Five flasks with 3 litre volume were prepared each with 4 g/l black tea (Lipton yellow label) steeped for 20 min. The flasks were given sugar sources and incolulants, as follows:
A. sucrose, 50 g/l; 10% inoculant I
B. fructose, 50 g/l; 10% inoculant I
C. glucose, 50 g/l; 10% inoculant I
D. sucrose 50 g/l; 17,5% inoculant II
E. sucrose 50 g/l; 10% inoculant III

The inoculant had been batch cycled in Narvik since December 2015, and had originally come from California, via a period cycling in Oslo. The most recent cycle in Narvik had been for 32 days. Inoculant I consisted 90% of liquid from the top of the flask and 10% from the middle of the flask; Inoculant II had 63% from top, 23% from middle and 14% from the bottom while Inoculant III had 75% from the middle and 25% from the bottom. The rationale for this differentiation was that the yeast dominates in the bottom and the *K. xylinus* in the more oxygen rich regions higher in the flask.

The progress of the fermentation was followed with optical density and spectrophotometric analyses using kits from Megazyme for sucrose, glucose, fructose, acetic acid, gluconic acid and ethanol.

Effectiveness of the culture

In contrast to the poor performance of a household-maintained culture reported by Sievers et al [3] this household-cycled culture showed efficient conversion of the sucrose to the end products, with the 50 g/l sucrose converted within 30 days and the monosaccharide concentrations peaking at 16 days. Both glucose and fructose were utilised, and the bacterial cellulose was produced also with fructose as starting substrate.



Progress of conversion of sucrose to intermediate and end products, Flask A.

Amount of bacterial cellulose produced, OD mg/litre/day

A	B	C	D	E
11,6	12,1	14,5	9,3	4,7

Conclusions

The household culture was shown to have retained a similar efficacy as that reported for a commercial culture [3]. Fructose is actively involved in the conversion, also being utilised for production of bacterial cellulose.

References

[1] Goh et al (2012) *Int. Food Res. J.* **19,** 153-158.
[2] Marsh et al (2014) *Food Microbiol.* **38,** 171-178.
[3] Sievers et al (1995) *System. Appl. Microbiol.* **18** 590-594.
[4] Greenwalt et al (1998) *Food Sci & Technol,* ***31*** *291-296.*