Acidic fermented beverages: Trends, functionality, production

Dr. Martin Senz
FIBW, Dept. Bioprocess Engineering and Applied Microbiology
Content

- Acidic fermented beverages
- Co-cultures and means of control of the fermentation process
- Functionality (technological; nutritional; …)
- Product characteristics and life style
- Market forecast and current activities
- Ingredients of kombucha and water kefir
- Current knowledge about health effects
- Home brewing and industrial production and possible hurdles
- Summary
Acidic fermented beverages

+ Mainly in co-culture fermented beverages based on diverse raw materials
+ Different symbiotic cultures of bacteria and yeast
+ Most relevant fermentations for food & beverages: Acidic-, lactic- and alcoholic fermentation

• Kombucha
• Ginger Beer
• Kefir / Water kefir
• Kvass
• *Berliner Weiße*
• *Lambic Beer*
• …
Examples: Kombucha / Water kefir

Traditionally produced via “Tea fungi”

- Sweetened *(Sucrose, 60-80 g/L)* Tea *(black, green)* with tea fungi and 10% old kombucha
- *SCOBY = Symbiotic Culture of Bacteria and Yeast*
- Mostly in big closed vessels with enough head space
- Incubation for 7 – 10 d, 18 °C to 26°C
- Characteristic sweet acetic taste, mild carbonized

Yeast → Usage of sucrose and production of alcohol

Oxygen

Bacteria *(Acetic acid and lactic acid bacteria; AAB and LAB)*
- Alcohol to acid (AAB)
- Formation of bacteria cellulose

Traditionally produced via “Kefir-crystals”

- Sweetened *(Sucrose, 60-80 g/L)* water with kefir crystals and dried fruits *(→ nitrogen source)*
- Mostly in big closed vessels with minor head space
- Incubation for 2 – 3 d, 18 °C to 26°C
- Characteristic sweet acetic, fresh and mild carbonized taste. Similarities to young fruit wine

Yeast → Usage of sucrose and production of alcohol

Bacteria (LAB, AAB)
- Acid production (lactic and acetic acid)
- Formation of Kefiran

VLB Berlin / Research Institute for Biotechnology and Water / Department Bioprocess Engineering and Applied Microbiology / Dr. Martin Senz
Co-culture fermentation control

Process parameters:

- Type and combination of starter cultures
- Quality of starter cultures
- Type and concentration of raw materials
- Concentration and adding regime of inoculation
- Temperature
- pH value
- Oxygen regime and redox potential

→ Are used to control the production process
→ Influences the functionality of the beverage
Impact on functionality: technological point of view

- Technological functionality
  - Aroma formation, blending of bad tastes
  - Shelf life/safety
  - Reproducibility/constant quality of the product
  - Integration of CO₂, freshness
  - Mouth feeling
  - Texture
  - Color
Impact on functionality: nutritional & physiological point of view

antibiotic, antioxidative, antihypertensive, cholesterol-lowering, stimulating ...

Secondary plant products (phenols, flavonoids, carotenoids, etc.)

Probiotic cultures
- Competitive exclusion of pathogens
- Altered metabolism/energy utilization
- Immune stimulation
- Treatment of infectious diarrhea

↑ digestion

↑ vitamins
↓ ANS

nutritional functionality

prebiotics
- Di- and oligo- and polysaccharides, inulin, lactulose, raffinose, fructane, etc.

www.isernhaeger.de
www.prophyta.de
www.davehubbleecology.blogspot.com
Further relevant properties of the beverage that impacts the buying behavior:

- Vegan, veggie, free of gelatin
- Free of gluten, lactose
- Kosher, halal (alcohol free)
- "Clean label" free of dye stuffs, preservatives, E-No. ...
- Bio, organic, raw
- Natural, fermented
- Regional, fair-trade...

Properties...

www.isernhaeger.de
www.davehubbleecology.blogspot.com
www.davehubbleecology.blogspot.com
... resp. the product character has to comply with the individual life style:

„Health style meets life style“

Further trends:

- Partly renaissance of traditional processing techniques (e.g. fermentation-workshops)
- Home brewing
- Diversity, individuality
- Raising impact of popular literature and social-media
„The rules of social media era apply also for water kefir. In case of the beverage it means: More important than the beverage that you drink is the picture you put on the Web to impose your followers“

(Frederik Eikmanns, Süddeutsche Zeitung)
Promising market forecasts

+ The Europe Kombucha market generated nearly $180 million for the year 2016. Europe Kombucha market is expected to progress with a healthy 25.25% CAGR for the forecast period of 2017-2025, generating a total of approximately $1365 million by the end of 2025 (INKWOOD Research, Market Research Report; www.inkwoodresearch.com/reports/europe-kombucha-market; Abrufdatum: 2.Okt.2018)


+ Global Kombucha market is expected to progress with 23.0% CAGR leading to a total of approximately USD 5.54 billion by the end of 2025. The global consumption for kombucha was estimated to be 82,960.9 kilo liters in 2016 (≡ ca. 4% of beer [~2 billion hl]). (Source: www.grandviewresearch.com/industry-analysis/kombucha-market; Abrufdatum: 2.Okt.2018)

+ Global kefir market is expected to progress with 7.3% CAGR till 2023. → Increase from 1.18 billion USD in 2016 to 1.93 billion USD in 2023. (www.reuters.com/brandfeatures/venture-capital/article?id=18632; Abrufdatum: 2.Okt.2018)
Current activities reflect the market forecasts

+ VLB: Increased request for corresponding product developments
+ Diversification of the product range of well known companies:
  
  2018… *Coca-Cola* buys the Australian kombucha producer *Organic & Raw Trading Co.* with the brand *Mojo*.  
  2018… *Molson Coors* takes over *Clearly Kombucha*.  
  2018… *Starbucks* enters the kombucha category via *Evolution Fresh* brand.  
  2016… PepsiCo takes over the kombucha giant *Ke-Vita*. 
Kombucha compounds

RAW MATERIALS

Polyphenols → antioxidant → detoxify

Caffeine → stimulating

SCOBY

KOMBUCHA

↑ Vitamins
B₁, B₂, B₆, B₁₂, C
Coenzymes

↑ Gluconic acid
→ Mild acid for the taste

↑ Acetic acid
→ Microbial control

↑ Glucuronic acid
→ Detoxification
→ Phenol transportation/
Antioxidant transportation and bioavailability

↓ Sucrose
→ Energy source for the microorganisms

↑ Ethanol
→ Present in low concentration
→ Prevents cardiovascular disease

Kombucha compounds

Many studies regarding (tea-) compounds and kombucha

Kombucha – SCOBY Microorganisms

+ **Main isolated yeasts:**

  - *Zygosaccharomyces bailii*
  - *Schizosaccharomyces pombe*
  - *Torulospora delbreuckii*
  - *Rhodotorula mucilaginosa*
  - *Brettanomyces bruxellensis*
  - *Candida stellata*
  - *Saccharomycodes ludwigii*
  - *Saccharomyces spec.*
  - *Pichia spec.*

+ **Main isolated bacteria:**

  - **Acetic acid bacteria**
    - *Gluconacetobacter xylinus*
    - *Gluconacetobacter kombuchae sp.*
    - *Acetobacter pasteurianus*
    - *Acetobacter aceti*
    - *Acetobacter intermedius*
    - *Gluconobacter oxydans*

  - **Lactic acid bacteria**
    - *Lactobacillus spec.*

---

KBI OSU DNA Sequence Study Analysis Report:
Study of the Oregon State University & Kombucha Brewers International, with nearly 100 sequenced samples of 70 participants

Bacteria profile of average KBI SCOBY

Fungal profile of average KBI SCOBY

www.kombuchabrewers.org/kbi-osu-dna-sequence-study-analysis-report/
Common declarations for kombucha: „probiotic cultures“

- Russian noblest Ilja Metchnikoff as founder of the probiotic term
  
  Thesis: LAB reduce putrefaction in the gut → altered composition of microbiota as therapy

- Definition probiotics according FAO/WHO, 2002:
  „live microorganisms which when administered in adequate amounts confer a health benefit on the host“.

- Probiotics: most relevant genera in 2015 Lactobacillus (59%), Bifidobacterium (31%) and Streptococcus (6%)¹

  → So far no established beneficial (probiotic) effects of acetic acid bacteria and the dominant yeast in kombucha!

Common declarations for kombucha: vitamins and organic acids – *own studies* -
Common declarations for kombucha: vitamins and organic acids

Data from scientific publications

<table>
<thead>
<tr>
<th>Substanz</th>
<th>g/L</th>
<th>g/L</th>
<th>Fermentationszeit (Tage)</th>
<th>Quelle</th>
</tr>
</thead>
<tbody>
<tr>
<td>Glucuronsäure</td>
<td>0,003</td>
<td>100</td>
<td>21</td>
<td>Loncar (2000)</td>
</tr>
<tr>
<td>Glucuronsäure</td>
<td>0,015</td>
<td>100</td>
<td>21</td>
<td>Loncar (2006)</td>
</tr>
<tr>
<td>Glucuronsäure</td>
<td>0,57</td>
<td>50</td>
<td>14</td>
<td>Talawat (2006)</td>
</tr>
<tr>
<td>Glucuronsäure</td>
<td>1,30</td>
<td>100</td>
<td>21</td>
<td>de Filippies (2018)</td>
</tr>
<tr>
<td>Glucuronsäure</td>
<td>1,69</td>
<td>100</td>
<td>9</td>
<td>Jayabalal (2007)</td>
</tr>
<tr>
<td>Glucuronsäure</td>
<td>1,71</td>
<td>100</td>
<td>18</td>
<td>Jayabalal (2007)</td>
</tr>
<tr>
<td>Gluconsäure</td>
<td>12,00</td>
<td>70</td>
<td>10</td>
<td>Blanc (1996)</td>
</tr>
<tr>
<td>Gluconsäure</td>
<td>31,00</td>
<td>70</td>
<td>25</td>
<td>Blanc (1996)</td>
</tr>
<tr>
<td>Gluconsäure</td>
<td>2,80</td>
<td>70</td>
<td>10</td>
<td>Sievers (1995)</td>
</tr>
<tr>
<td>Gluconsäure</td>
<td>5,00</td>
<td>70</td>
<td>20</td>
<td>Sievers (1995)</td>
</tr>
<tr>
<td>Gluconsäure</td>
<td>8,90</td>
<td>70</td>
<td>30</td>
<td>Sievers (1995)</td>
</tr>
<tr>
<td>Gluconsäure</td>
<td>2,44</td>
<td>unknown</td>
<td>7</td>
<td>Chakravorty (2016)</td>
</tr>
<tr>
<td>Gluconsäure</td>
<td>6,38</td>
<td>unknown</td>
<td>14</td>
<td>Chakravorty (2016)</td>
</tr>
<tr>
<td>Gluconsäure</td>
<td>7,36</td>
<td>unknown</td>
<td>21</td>
<td>Chakravorty (2016)</td>
</tr>
<tr>
<td>Gluconsäure</td>
<td>1,20</td>
<td>unknown</td>
<td>unknown</td>
<td>Pietschmann et al. (1996)</td>
</tr>
<tr>
<td>Gluconsäure</td>
<td>2,30</td>
<td>100</td>
<td>21</td>
<td>de Filippies (2018)</td>
</tr>
<tr>
<td>Gluconsäure</td>
<td>39,00</td>
<td>100</td>
<td>60</td>
<td>Chen and Liu (2000)</td>
</tr>
<tr>
<td>Ethanol</td>
<td>0,28</td>
<td>unknown</td>
<td>7</td>
<td>Chakravorty (2016)</td>
</tr>
<tr>
<td>Ethanol</td>
<td>0,14</td>
<td>unknown</td>
<td>14</td>
<td>Chakravorty (2016)</td>
</tr>
<tr>
<td>Ethanol</td>
<td>0,07</td>
<td>unknown</td>
<td>21</td>
<td>Chakravorty (2016)</td>
</tr>
<tr>
<td>Ethanol</td>
<td>4,07</td>
<td>70</td>
<td>7</td>
<td>Velicanski (2013)</td>
</tr>
<tr>
<td>Ethanol</td>
<td>5,50</td>
<td>100</td>
<td>20</td>
<td>Chen and Liu (2000)</td>
</tr>
<tr>
<td>Ethanol</td>
<td>3,60</td>
<td>70</td>
<td>10</td>
<td>Sievers (1995)</td>
</tr>
<tr>
<td>Ethanol</td>
<td>7,00</td>
<td>70</td>
<td>30</td>
<td>Sievers (1995)</td>
</tr>
</tbody>
</table>

Recommended daily doses of Vitamin B12:

3 µg/day (DGE Reference value)

Comparison:
- 1 Liter milk ~4µg
- 50g Edamer ~1 µg
- 100g calves ‘liver ~60µg
Microorganisms in water kefir

**Yeast:**
- *Kluyveromyces marxianus*
- *Candida kefyr*
- *Kluyveromyces lactis* var. *Lactis*
- *Debaryomyces hansenii*
- *Dekkera* spec.

**Bacteria:**
- **Lactic acid bacteria**
  - *L. kefiri*, *L. kefiranofaciens*, *L. casei*, *L. paracasei*, *L. parakefiri*, *L. plantarum*, *L. acidophilus*, *L. amylovorus*, *L. brevis*, *L. buchneri*, *L. crispatus*, *L. delbrueckii*,
  - *L. salivarius*, *L. sunkii*, *Lc. garvieae*, *Lc. lactis*, *Leuconostoc mesenteroides*, *O. oeni*, *Pediococcus* sp.,
  - *Tetragenococcus* halophilus

- **Acetic acid bacteria**
  - *Komagataeibacter hansenii*,
  - *Zymomonas* spec.
Traditional vs. industrial production

+ Traditionally produced in small scale:
  private, pharmacies, …
+ Reuse of the SCOBY
+ Limited control:
  - Unknown age of the cultures
  - Composition of the SCOBY is unknown and not constant
  - Risk of mold formation
  - Incomplete process → high alcohol
    to long process → vinegar

+ Industrial process should be done under quality assurance under quality management and control aspects:
  + Process knowledge essential
  + Reproducible product properties
  + Control points mandatory
+ Application of defined starter cultures
+ Control of process and parameter (if possible online)
Process knowledge is the basis to meet the challenges of desired product variations

E.g.:
+ Enrichment with metabolites (vitamins, org. acids, …)
+ Defined sugar profile after fermentation
+ Enrichment with beneficial microorganisms
+ Alcohol concentration and its stability
+ Raw products
+ Standardization and classification of acidic fermented beverages
+ …
Summary

+ Steady growth of co-culture fermented acidic beverages that is accompanied with the increasing healthy life style of the population

+ Many possibilities to bring functionality with innovative character into the beverage

+ For producers: The desired product properties has to be harmonized with the production conditions

+ Growing market of diverse fermented products → Need for standardization and classification

+ Latter can contribute to have products with correct and clear declarations on the market
Thank you very much!

VLB Berlin

Dr.-Ing. Martin Senz

Bioprocess Engineering and Applied Microbiology (BEAM)
Research Institute for Biotechnology and Water (FIBW)
m.senz@vlb-berlin.org
www.vlb-berlin.org
References

+ Pietschmann, M. u.a., Gärgetränke des Handels - Zusammensetzung, ernährungsphysiologische und rechtliche Beurteilung, Deutsche Lebensmittel-Rundschau, 96 (6), 203 - 210, 2000
+ Nguyen, N. K., Nguyen, P. B., Nguyen, H. T., & Le, P. H. (2015). Screening the optimal ratio of symbiosis between isolated yeast and acetic acid bacteria strain from traditional kombucha for high-level production of glucuronic acid. LWT-Food Science and Technology, 64(2), 1149-1155.
Video: Water-kefir fermentation