VI NEGAR MAKING

Acetic acid
Vinegar

- Legally means acetic acid product made from apples (like wine is from grapes)
- To be legal, vinegars in US must contain a minimum of 4% of acetic acid
Different countries - different starting materials

- Continental Europe - grape wines
- Great Britain - malt
- Hawaii and Far East - pineapple
- USA - apple
Alcoholic Fermentation

Alcohol

O₂

VINEGAR
IMITATION VINEGAR

- In late 1800s chemist learned how to make acetic acid
- Manufacturers added water to reduce strength to 5%, colored it and sold it as a vinegar
- Imitation vinegar is still manufactured and by law the label must state that it is diluted acetic acid
Acetic acid (fermentation?)

- $2\text{CH}_3\text{CH}_2\text{OH} + \text{O}_2 = 2\text{CH}_3\text{COOH} + 2\text{H}_2\text{O}$

- Ethanol 92g/mol
- Acetic acid 120 g/mol
- Theoretical yield $(120/96) \times 100 = 130\%$
- Practical yield $\sim 120\%$
Acetic acid bacteria

- obligatory aerobic
- nitrogen-fixing bacteria
- known for producing acid as a result of metabolic processes
- *Acetobacter aceti*
FERMENTED VINEGAR

- Made from good alcohol (wine, beer) that is fermented using acetic bacteria that convert alcohol to acetic acid
- Process can be controlled by using great care in cleanliness and introducing chosen yeast (wine) and bacteria (vinegar) to obtain good quality every time
Methods of making vinegar

- **SLOW**
  - Home making
  - Orleans process

- **RAPID**
  - The generator system
  - Submerged fermentation system (bubble method)

Better flavor of vinegar
WINE VINEGAR

- Wine that usually contains 11-12% alcohol must be diluted to 5.5-7% alcohol before using it to make vinegar.
- Vinegar should contain at least 5% acetic acid to as required for preserving and pickling.
MAKING WINE VINEGAR

- **Utensils**
  - Use glass, stainless still, enamel. Vinegar leaches molecules from iron and aluminum.

- **Sanitize utensils**
  - Everything that will touch vinegar should be sanitized. Soak everything for 20 minutes in a solution of 2 tablespoons chlorine laundry bleach to 1 gallon of water.
MAKING VINEGAR

- Dilute wine to 5.5-7% alcohol with water
- Fill sterilized containers with about 2/3 full
- Add bacteria cultures
  - Leaving wine exposed to air may sometimes start the process, but is very risky because some other organisms may grow
  - It is recommended to order pure cultures of bacteria sometimes called mother of vinegar
  - They can be purchased in any store that supplies wine making equipment. It usually comes like a clear liquid in the jar
Making Vinegar

- Cover the container with a cloth to keep the insects out while allowing air to freely reach the stock.
- Two factors require special attention when making vinegar at home:
  - Oxygen supply
  - Temperature
TEMPERATURE

- Between 80-85 F is ideal
- Low or fluctuating temperatures slow the process
- High temperatures will kill bacteria
FORMATION OF THE FILM

- After some time an acetic acid film called “mother” will form.
- It should not be disturbed.
- It often becomes heavy enough to fall, in such case remove it and discard it.
VINEGAR

- Living bacteria are in the liquid
- They can be used to start the new batch of vinegar so it is not necessary to purchase new starter bacteria
- Full fermentation will take 3-4 weeks
PRESERVING VINEGAR

- Filter vinegar through a layer of cheese cloth to remove formed film—mother of vinegar, before pasteurization.
- Heat vinegar before pouring it into sterilized bottles.
- Bottle and place in hot water bath.
- In both cases the temperature of the vinegar must reach at least 140F and should not exceed 160F and should be held at that temperature for at least 30 minutes.
- Stored vinegar will stay in excellent condition almost indefinitely if it is pasteurized.
AGING VINEGAR

- Vinegar has a strong, sharp bite when it is just made.
- It becomes mellow when aged.
- It usually lasts 6 months or longer when stored at cool, steady temperature (50-60 F).
- This undisturbed rest also allows suspended solids to fall, making the vinegar clear and bright.
- Once vinegar is ready it should be kept away from oxygen because acetic acid could be converted into water and carbon dioxide.
FLAVORED VINEGAR

- Flavoring can be added to homemade vinegar just before bottling.
- Some additives can include: garlic, ginger, or any combination of dried fresh herbs.
- Place flavoring material in a small cheesecloth bag and suspend in vinegar until the desired strength is reached.
- It will take approximately 4 days, except for garlic which takes only 1 day.
Orleans process

- Wine or cider is fermented in wooden barrels or covered vats.
- There are screened air vents located in the top part of the container.
- Vents are screened to prevent Drosophila.
- A starter culture is put into the container, and the fermentation is continued until the acetification is complete.
- About 3/4 of the vinegar will be drawn off and replaced with fresh wine or other fermentable liquid.
- The process will keep repeating itself as long as quality vinegar can still be produced.
- This process is very slow (several weeks) and is usually done in small batches.
Orleans process
Rapid-generator process
(Schutzenbach)

- Fermentation is done in a container that consists of two chambers.
- The larger (upper) chamber is packed with solid materials almost to the top (wood shavings, corncobs etc.) (bacteria attach to it)
- The upper chamber is separated from the lower chamber by a screen.
- Air is injected & blown upward through the screen and through the solid materials, & the air escapes through the top.
- The fermenting liquids are distributed evenly over the top of the material, & allowed to percolate through the material.
- The resulting liquid is then pumped back to the top & recirculated until the alcohol content is reduced to 1/2 percent.
- The vinegar is drawn off and fresh alcoholic solution is added.
Frings acetator or generator
Submerged culture fermentation (bubbling)

- First was developed during World War II for the production of penicillin.
- Tiny bubbles of air are passed through the solution of ethanol.
- Agitated by propellers, dispersing cells of acetic acid bacteria.
Essentials for vinegar generator

- Must provide for the introduction of fresh substrate without disturbing the content of alcohol or the surface growth
- Must provide easy sampling for titration
- Must protect system from contamination
- Must have means of temperature control
Rate of acetic acid production

- The inherent capacity of *Acetobacter aceti* to convert alcohol to acetic acid
- The amount of ethyl alcohol (6-8% opt., 12% tolerated)
- The temperature (80°F opt., 68-96°F range)
- The amount of aeration of surface area of the growth material
LACTIC ACID
FERMENTATION
Requirements

- Starting fermentable sugar (glucose, lactose)

- Microorganisms (lactic acid bacteria or fungi)
Fermentation

(a) LACTIC ACID FERMENTATION

Glucose → Glycolysis → Pyruvic acid

Lactic acid

NAD+ → NADH + H+

Diagram showing the process of lactic acid fermentation with glucose, glycolysis, pyruvic acid, lactic acid, NAD+, and NADH + H+.
Microorganisms

- Lactic acid fermentation is caused by some fungi and bacteria.
- The most important lactic acid producing bacteria is *Lactobacillus*.
- Other bacteria which produce lactic acid include:
  - *leuconostoc mesenteroides*,
  - *pediococcus cerevisiae*,
  - *streptococcus lactis*,
  - *bifidobacterium bifidus*. 
Lactic acid bacteria refers to a large group of beneficial bacteria that have similar properties and all produce lactic acid as an end product of the fermentation process.

They are widespread in nature and are also found in our digestive systems.
Lactic acid bacteria

- **Homolactic fermentation**
  - The fermentation of 1 mole of glucose yields two moles of lactic acid;

- **Heterolactic fermentation**
  - The fermentation of 1 mole of glucose yields 1 mole each of lactic acid, ethanol and carbon dioxide;
Nisin

- Nisin was the first bacteriocin derived from fermentation of a lactic-acid bacterium.
- Approved by the FDA in April 1989 to prevent the growth of botulism spores in pasteurized process-cheese spreads.
- Does not inhibit Gram-negative organisms, yeasts or fungi, but does inhibit most Gram-positive organisms including spore-formers such as Clostridia botulinum and heat-resistant spoilage organisms.
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<tr>
<th>Homofermenter</th>
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<th>Obligate heterofermenter</th>
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<tr>
<td><em>Enterococcus faecium</em></td>
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<td><em>Lactobacillus curvatus</em></td>
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<td><em>Streptococcus bovis</em></td>
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<td><em>Leuconostoc dextranicum</em></td>
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<td><em>Pediococcus acidilactici</em></td>
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<td><em>Pedicoccus damnosus</em></td>
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<td><em>Pediococcus pentocacus</em></td>
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Conditions for lactic acid fermentation

- Addition of a sufficient amount of fermentable carbohydrates
- Reduced $O_2$ during the fermentation process and storage of the fermented product.
- Rapid multiplication of the starter culture and sufficient production of lactic acid.
Lactic fermentation products

- Western world: yogurt, sourdough breads, sauerkraut, cucumber pickles and olives
- Fermented meats
- Middle East: pickled vegetables
- Korea: kimchi (fermented mixture of Chinese cabbage, radishes, red pepper, garlic and ginger)
- Russia: kefir
- Egypt: laban rayab and laban zeer (fermented milks), kishk (fermented cereal and milk mixture)
- Nigeria: gari (fermented cassava)
- South Africa: magou (fermented maize porridge)
- Thailand: nham (fermented fresh pork)
- Philippines: balao balao (fermented rice and shrimp mixture)
Fermented vegetables

- Sauerkraut
- Pickles
Fermentation

- Lactic acid fermentations are carried out under three basic types of condition:
  - dry salted,
  - brined and
  - non-salted.
- Salting provides a suitable environment for lactic acid bacteria to grow which impart the acid flavor to the vegetable.
How does salt preserves food?

- The chloride ion is a bacterial poison
- Limits moisture availability
- Oxygen solubility is reduced
- Dehydrates protoplasm causing plasmolysis
- Interferes with enzyme action

- Generally yeast, bacteria and molds do not grow in saturated salt solution @ 26.5% sodium chloride at room temperature
Dry salted

- Vegetable is treated with dry salt
- The salt extracts the juice from the vegetable and creates the brine
- As soon as the brine is formed, fermentation starts and bubbles of carbon dioxide begin to appear.
- Fermentation takes between one and four weeks depending on the ambient temperature
Sauerkraut

- sauerkraut literally translates as acid cabbage
- *Leuconostoc mesenteroides*
- *Lactobacillus plantarum*
Shredded cabbage is placed in a jar and salt is added. Mechanical pressure is applied to the cabbage to expel the juice, which contains fermentable sugars and other nutrients suitable for microbial activity. The first micro-organisms to start acting are the gas-producing coccii (*L. Mesenteroides*). These microbes produce acids.

When the acidity reaches 0.25 to 0.3% (calculated as lactic acid), these bacteria slow down and begin to die off, although their enzymes continue to function. The activity initiated by the *L. mesenteroides* is continued by the lactobacilli (*L. plantarum* and *L. Cucumeris*) until an acidity level of 1.5 to 2% is attained.

The high salt concentration and low temperature inhibit these bacteria to some extent. Finally, *L. pentoaceticus* continues the fermentation, bringing the acidity to 2 to 2.5% thus completing the fermentation.
Temperature effect

- The optimum temperature for sauerkraut fermentation is around 21°C.
- A variation of just a few degrees from this temperature alters the activity of the microbial process and affects the quality of the final product.
- Therefore, temperature control is one of the most important factors in the sauerkraut process.
- A temperature of 18° to 22° C is most desirable for initiating fermentation since this is the optimum temperature range for the growth and metabolism of *L. mesenteroides*. 
**Effects of salt**

- Imparts firmness
- Inhibits putrefactive bacteria formation
- Withdraws water from the cabbage
- Added to a final concentration of 2.0 to 2.5%
Spoilage

- Aerobic soil micro-organisms break down the protein and produce undesirable flavor and texture changes.
- Dark colored sauerkraut (caused by spoilage organisms).
- Pink kraut is a spoilage problem. It is caused by a group of yeasts which produce an intense red pigment in the juice and on the surface of the cabbage.
Brine salted fermented vegetables

- For vegetables which inherently contain less moisture.
- A brine solution is prepared by dissolving salt in water (a 15 to 20% salt solution).
- Fermentation takes place well in a brine of about 20 salometer.
- The vegetable is immersed in the brine and allowed to ferment.
- The strong brine solution draws sugar and water out of the vegetable, which decreases the salt concentration.
- It is crucial that the salt concentration does not fall below 12%, otherwise conditions do not allow for fermentation. To achieve this, extra salt is added periodically to the brine mixture.
Pickles

- The washed cucumbers are placed in large tanks and salt brine (15 to 20%) is added.
- The cucumbers are submerged in the brine, ensuring that none float on the surface - this is essential to prevent spoilage.
- The strong brine draws the sugar and water out of the cucumbers, which simultaneously reduces the salinity of the solution.
- In order to maintain a salt solution so that fermentation can take place, more salt has to be added to the brine solution.
- If the concentration of salt falls below 12%, it will result in spoilage of the pickles through putrefaction and softening.
The color of the cucumber surface changes from bright green to a dark olive green as acids interact with the chlorophyll.

The interior of the cucumber changes from white to a waxy translucent shade as air is forced out of the cells.

The specific gravity of the cucumbers also increases as a result of the gradual absorption of salt and they begin to sink in the brine rather than floating on the surface.