FOOD BIOTECHNOLOGY IN NEPAL

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Food Biotechnology

- Broadly covers any use or improvement of living microorganisms to make food
- Today, this is divided into modern and traditional biotechnology
- Most applications of modern biotechnology to date involve plant raw material and ingredient production
- Whereas food bio-processing is still mainly traditional

Food Biotechnolgy

- Roots of biotechnology dates back some 12,000 years or so to the time of the agricultural revolution, when the first farmers learned how to select the best seeds, grow plants and cultivate cereal crops

- Modern biotechnology now allows food producers to do the same thing but with greater understanding, higher control and better selectivity

- The 21st century will bring another agricultural revolution first in increased crop yields and better raw materials, and second in a much reduced need for pesticides, fungicides, insecticides and herbicides

Food Biotechnology: Application

- Yeasts used in baking are optimally active around 30°C
- But these yeasts retain a low residual activity at refrigerated temperatures
- Isolation and greater screening of naturally occurring yeasts has shown new yeast strains which are still active below 12°C
- Such yeasts are now used commercially in bread and pizza doughs for refrigerate products
- These yeasts are self stable for weeks at 5-8°C; which will regain its full activity once the product is warmed to room temperature

Food Biotechnology: Application

- Rennet is an enzyme used in cheese making
- It is taken from the stomach lining of calves after slaughter
- Copying the gene responsible for producing rennet and inserting it into bacteria now means that rennet can be produced by fermentation
- Almost 50% of rennet comes out of industrial fermenters giving a more consistent and pure source of the enzyme
- Some *Mucor* spp produce microbial rennet which can be isolated from our traditional Murcha starters
- Modern food biotechnology can benefit not only the industrialized world but also developing countries

Food Biotechnology: Benefits

- Better testing tomatoes all year round. The fruit softens slowly which allows to pick tomatoes later, gaining added flavour and colour
- This may also apply to melons, peaches, bananas, and strawberries
- Healthier cooking oils. Corn, soybean, and other plants will be modified to lower the saturated fat content of cooking oils derived from these crops
- Potatoes that resist insects. New varieties will produce natural repellers against destructive insects
- Fruits and vegetables with higher levels of certain nutrients like vitamin C and beta-carotene
- The natural anti-oxidants may help reduce risk of chronic diseases such as some cancers and heart diseases

Food Biotechnology: Benefits

- Lower fat french fries and potato crisps. Higher starch potatoes will mean less fat absorption during frying
- Rice with higher levels of amino acids, lysine may help reduce childhood blindness due to lysine deficiency
- Eliminating allergy causing proteins from foods. Applied to rice, this will help Asian nations, which have high rates of rice induced food allergy
- Better methods to identify toxins, pathogens, contaminants in foods and raw materials
- Biotechnology will bring speed, accuracy and efficiency to modern food safety control
- Drought and flood tolerance agricultural system
- Heat and cold tolerance

Means of Fermentation

- Preserves perishable animal and plant products e.g. Sauerkraut, Pickle (Lactic acid bacteria), yogurt
- Reduces volume of material: Help transportation of raw foods- e.g. Cheese preservation by lactic acid bacteria and some mold
- Destroys undesirable factors in raw product: Tempeh fermentation by *Rhizopur oligosporus*, beany flavours and flatulence destroyed
- Enhances nutritive value: Vitamin B12 synthesized in Tempeh, PER improved in cereals, due to increase in Lysine

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- Improves appearance and flavour of some foods – e.g. Shoyu complements bland foods like rice and tofu, changes appearance and texture (solid to liquid), Angkak (rice moulded with Monascus) imparts red pigments to colour certain foods: sufu, rice, or meat

- Salvages some products that are otherwise unused. In the far east, fish pastes and sauces are made from fermentation of small fish and sea animals used as a flavouring or seasoning agents similar to shoyu

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- Reduces amount of energy in cooking Tempeh, Tarhana, Sufu
- Makes the product enjoyable. Besides its nutritional value, beer is a food that has been liked by people for centuries and certainly still be liked in the future
- Provides a safe product due to acid or alcohol content in the product. Wine has been consumed for safety reasons because water was badly contaminated with bacteria

Need for more research

- As population increases, more and more people will depend upon foods of plant origin including protein requirements

- Investigation on indigenous food fermentation is important to provide employment generating activities or developing a small scale enterprises at rural areas

Study Approach in Food Fermentation System

- Investigating the process: Identify processing steps. It is imperative to have someone who is familiar with food to judge whether or not the investigated product agrees with the native smell, flavour and appearances

- Characterizing the microorganisms
  - In-process investigation of microbes preferred
  - Microbes get killed in final product- e.g. Sufu (Actinomucor elegans)

- Isolate the useful microorganisms

- Purify, identify, and preserve organisms in culture collection

Fermentation

- Establish time and temperature relationship for optimum fermentation
- Disposal of by-products
  - Shoyu: Soybean press cake used as animal feed
  - Tempeh: Dehulled Soybeans soaked and cooked considerable amount of nutrient like soluble nitrogen and carbohydrate- cause water pollution
- Whether undesirable products formed
- High level of salt, low pH fairly safe
- Neutral pH and anaerobic condition- hazardous

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- Yield rate
  - involves determining actual dry weight losses. As microbes grow, there are metabolic losses with CO_2 and water as end products. Also the fermentation will go so fast that large amounts of heat are produced. Therefore, temperature control is essential

- Keeping quality
  - the shelf-life of the product should be determined, i.e., how long and at what temp. will the product remain in an edible state.

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- Biochemical changes
  - Changes in protein, polysaccharides, and lipids in the substrates. In soybean fermentation, soluble nitrogen, reducing substances and free fatty acids increase steadily
  - Total nitrogen, and lipid remain fairly constant
  - Often but not always, there will be changes in pH which may reverse direction. When fermentation of Tempeh is not stopped at proper time, the protease enzyme will form ammonia

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- Flavour constituents:
  - Flavour constituents are important in the acceptability of the product. This is rather complex process which occurs due to metabolic fate of responsible microorganisms on carbohydrates, proteins and lipids in tempeh fermentation, beany flavour of soybean disappears.
- Enzymes formed by microorganisms- complex area of study and requires years of investigations

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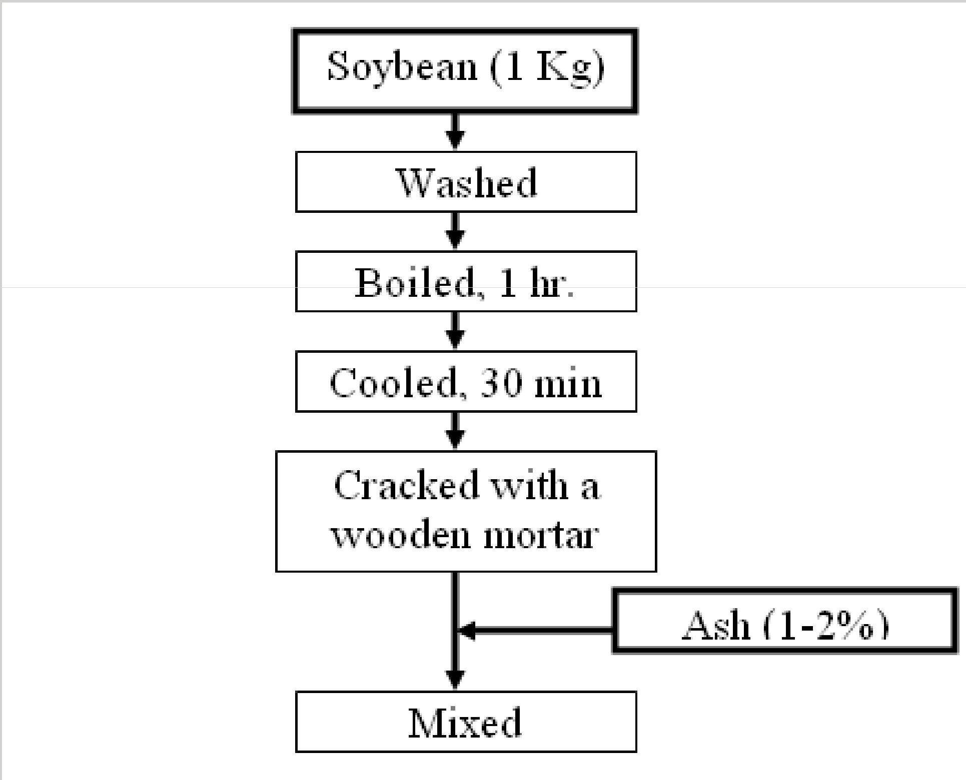
- The final product should be examined for compounds, not present in the original material. When microorganisms whether bacteria, yeast or fungi grow in any substrate, one can expect to find new compounds. Some of the new compounds are important for nutritive value of the product. In tempeh fermentation, vitamins (Niacin, and riboflavin) are often produced in large amounts.

Improvement

- Fermentation Process
- Microorganisms
- Designing Equipment
- Scale-up of the Process
- Packaging
- Shelf-life
- Marketing

Distribution of soybean foods in Asia

Traditional Process of Kinema Observed in Dharan



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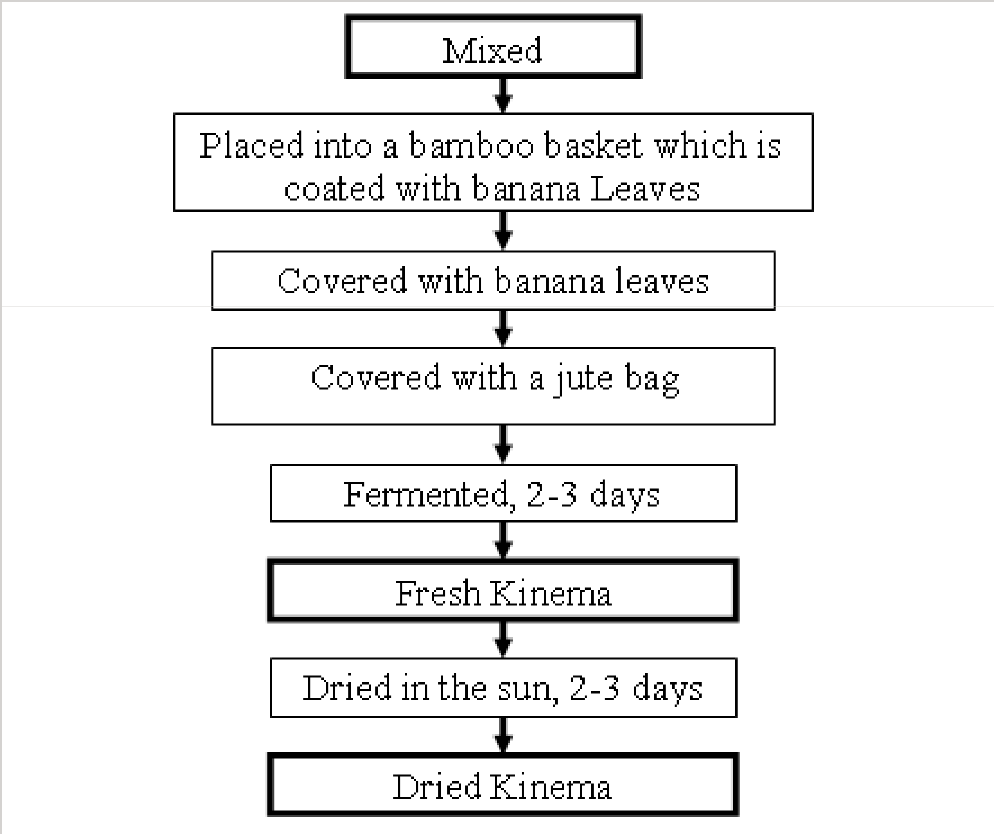
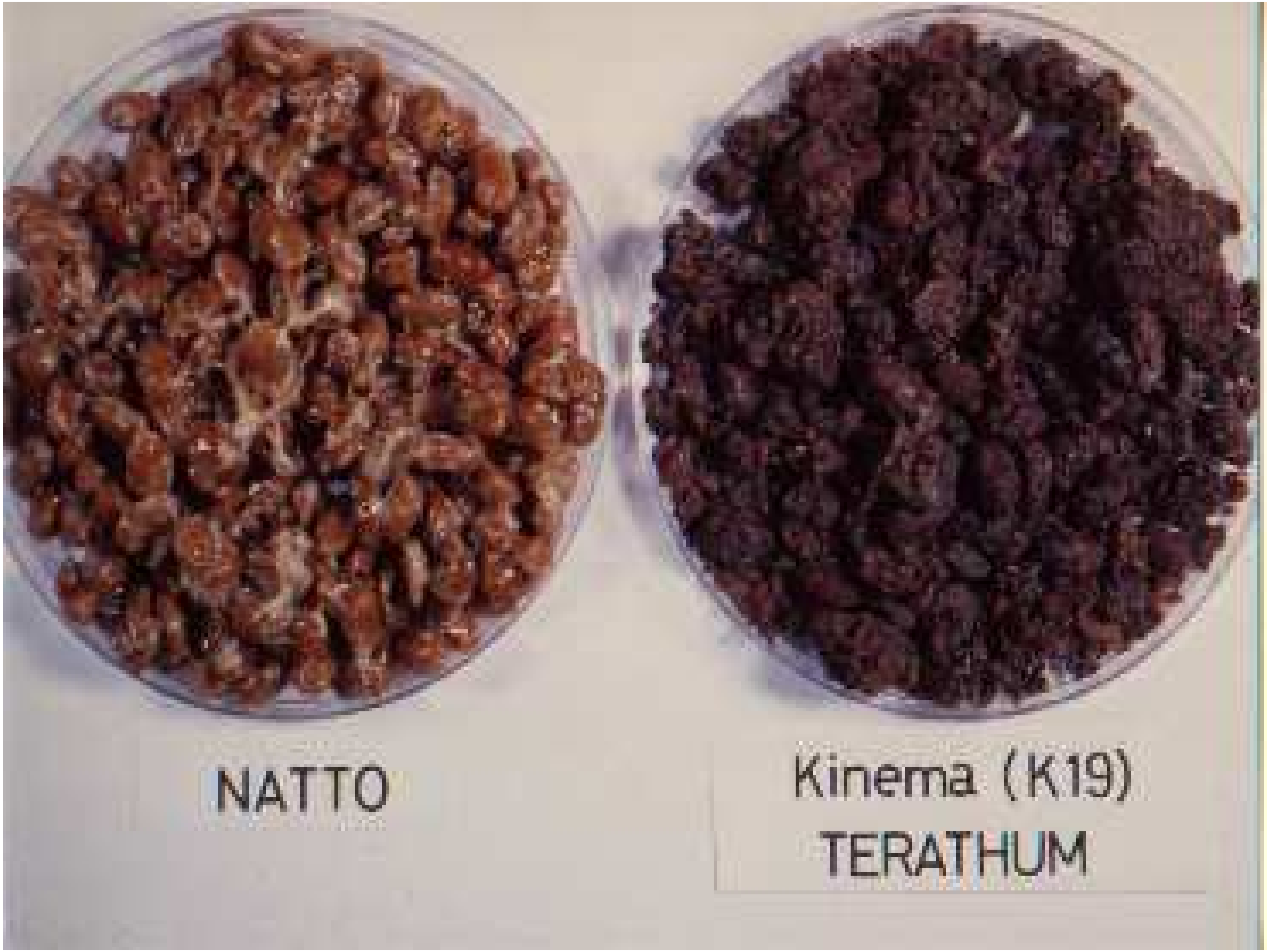
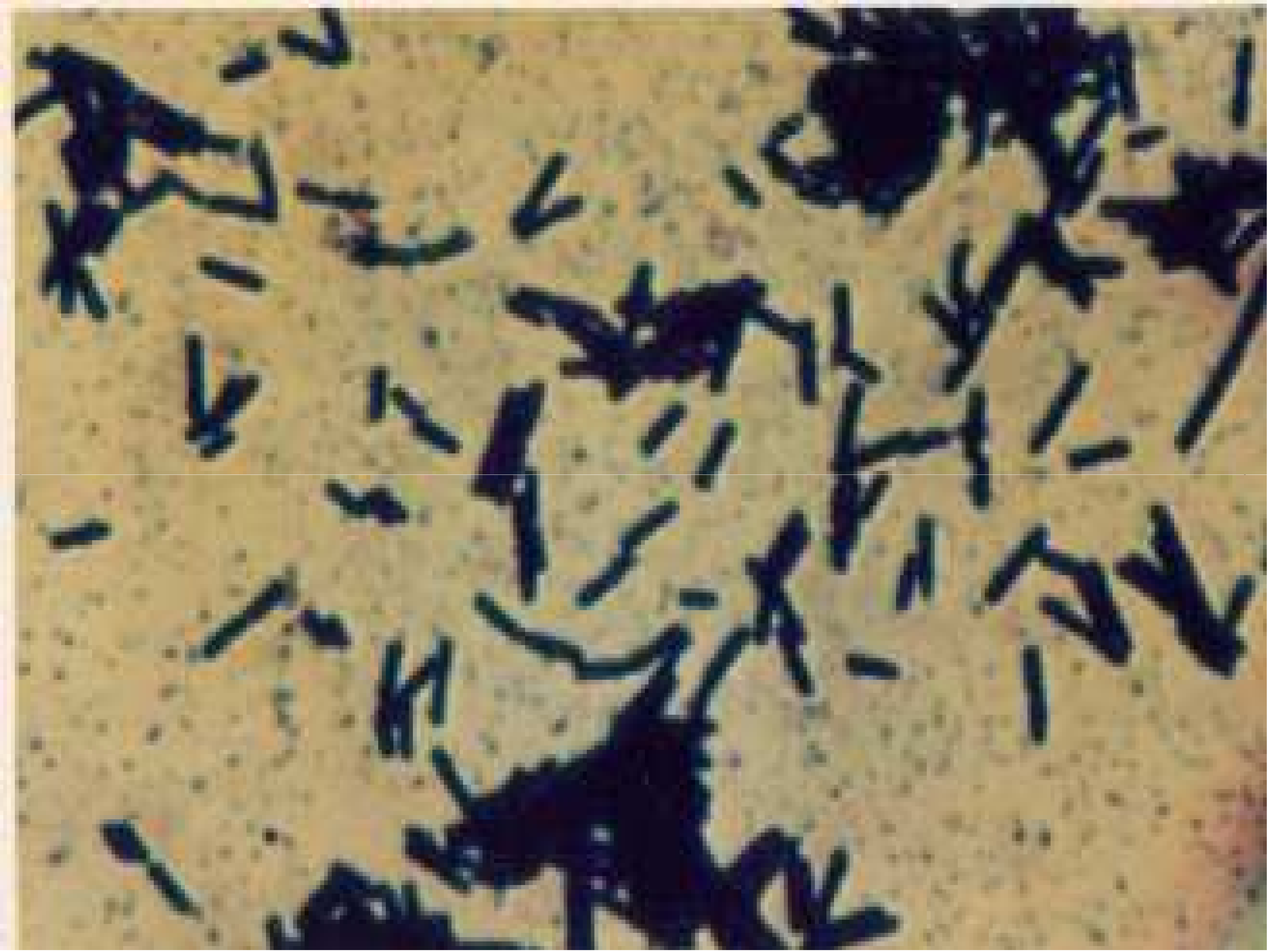

Mixed
Placed into a bamboo basket which is coated with banana Leaves
Covered with banana leaves
Covered with a jute bag
Fermented, 2-3 days
Fresh Kinema
Dried in the sun, 2-3 days
Dried Kinema

Brown soy bean (K19)
TERATHUM
Brown soy bean (K22)
TERATHUM
White soy bean
DHANKUTA


NATTO
Kinema (K19)
TERATHUM



Nutritional Value of Natto

Parameters	Per 100 gm. Natto
Energy	191.0 K Cal
Water	58.5 g
Protein	16.5 g
Fat	10.0 g
Non-fibrous	2.3 g
Carbohydrate	2.6 g
Ash	92.0 mg
Ca	0.07 mg
Vitamin B1	0.56 mg
Vitamin B2	0.56 mg
Nicotinic acid	1.1 mg

Enzymes of B. subtilis (Natto)

- Bacillus excretes enzymes:
  - Proteinase
  - Amylase
  - Mannase
  - Cellulase
  - catalase

Medicinal Effect of Natto

- Antibiotics:
  - Dipicolinic acid produced by B. subtilis, prevented growth of underirable microorganisms.
- Elimination of Microflora in Intestine:
  - B. subtilis after oral administration, eliminated pathogenic bacteria from intestinal canal. Tablets from lyophilized powder of B. subtilis var natto after their effect on enterobacterial flora after oral administration to humans and animals.

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- Anticancer Activity:
  - B. subtilis (natto) culture filtrate was cytolytic toward Ehrlich ascites carcinoma cells.

- Blood Pressure Depression:
  - Significant depression of blood pressure was observed by intake of natto, natto juice or an alcoholic extract of natto. The blood pressure of subjects fed natto soup was 20 to 30 mm Hg less than of control.

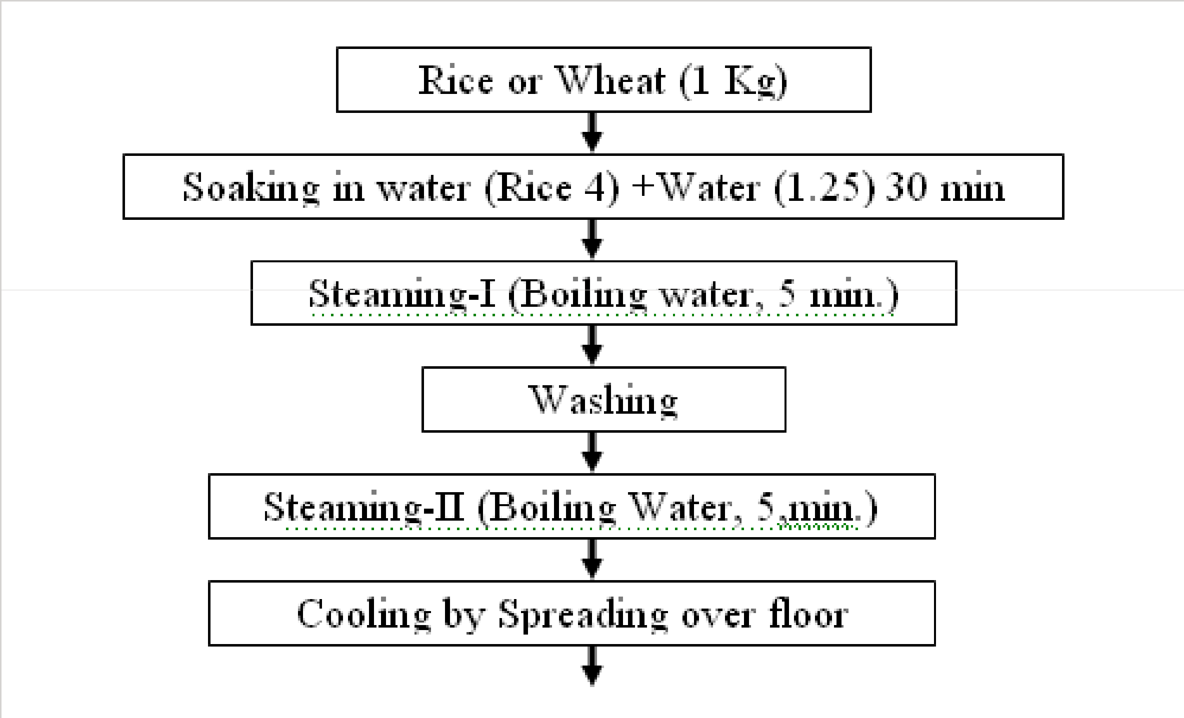
Name of Starter in different Countries

Name	Country
Bubod	Philippines
Chu	China
Luk-Paeng	Thailand
Nurook	Korea
Ragi	Indonesia
Ragi	Malaysia
Murcha	Nepal
Murcha	India
Bara-Koji	Japan

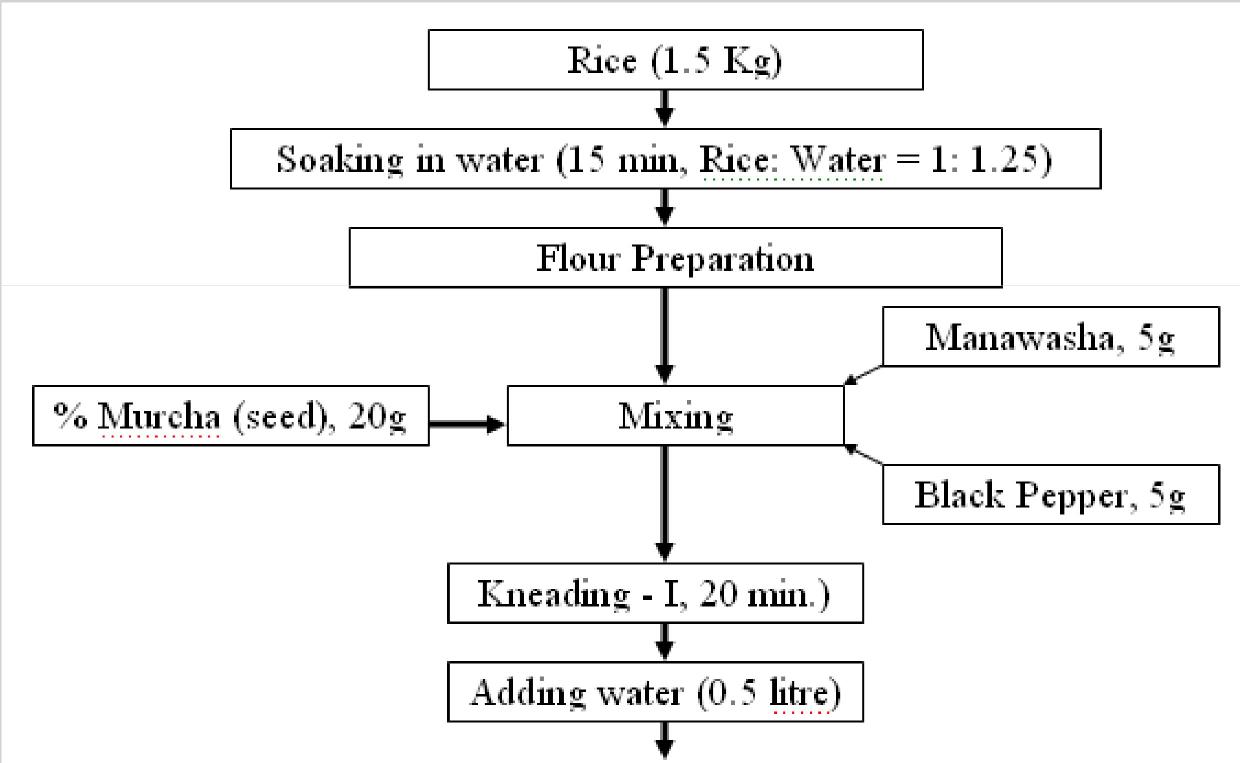
Rice Wine

Name of rice wine	Country
Brem Bali	Indonesia
Mie-chiu	China
Sake	Japan
Sato	Thailand
Sonti	India
Tapoi	Philippines
Yakju	Korea
Jand, Raksi	Nepal

Traditional Method of Manufacturing Mana



Traditional Manufacturing Process of Manapu



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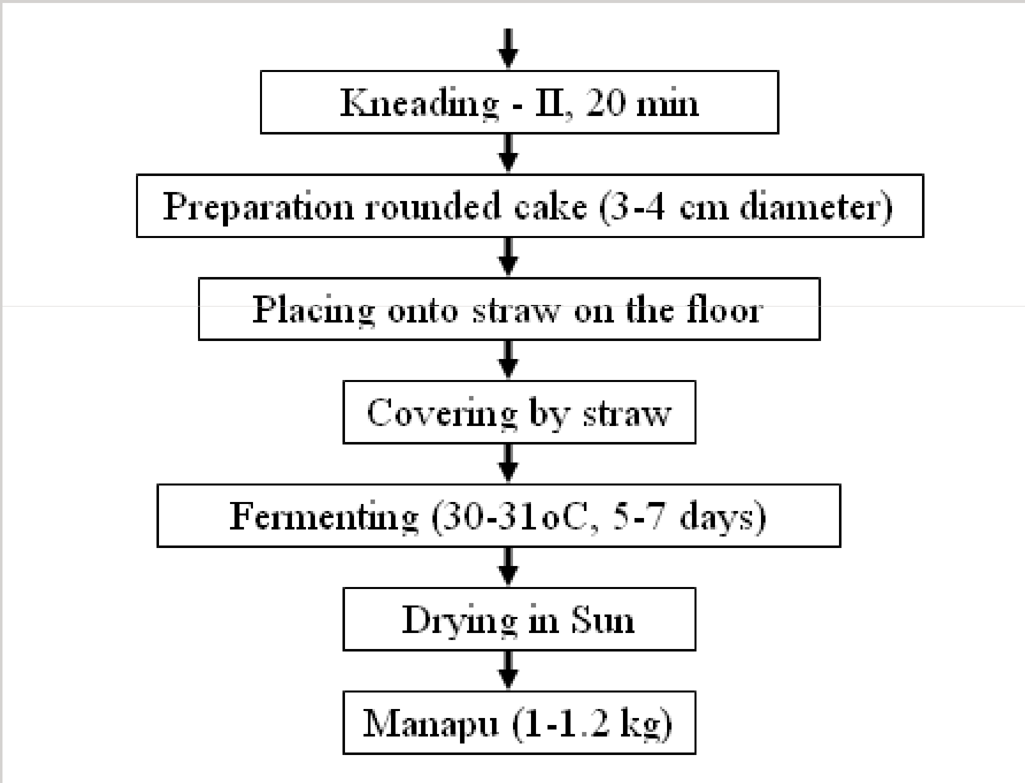
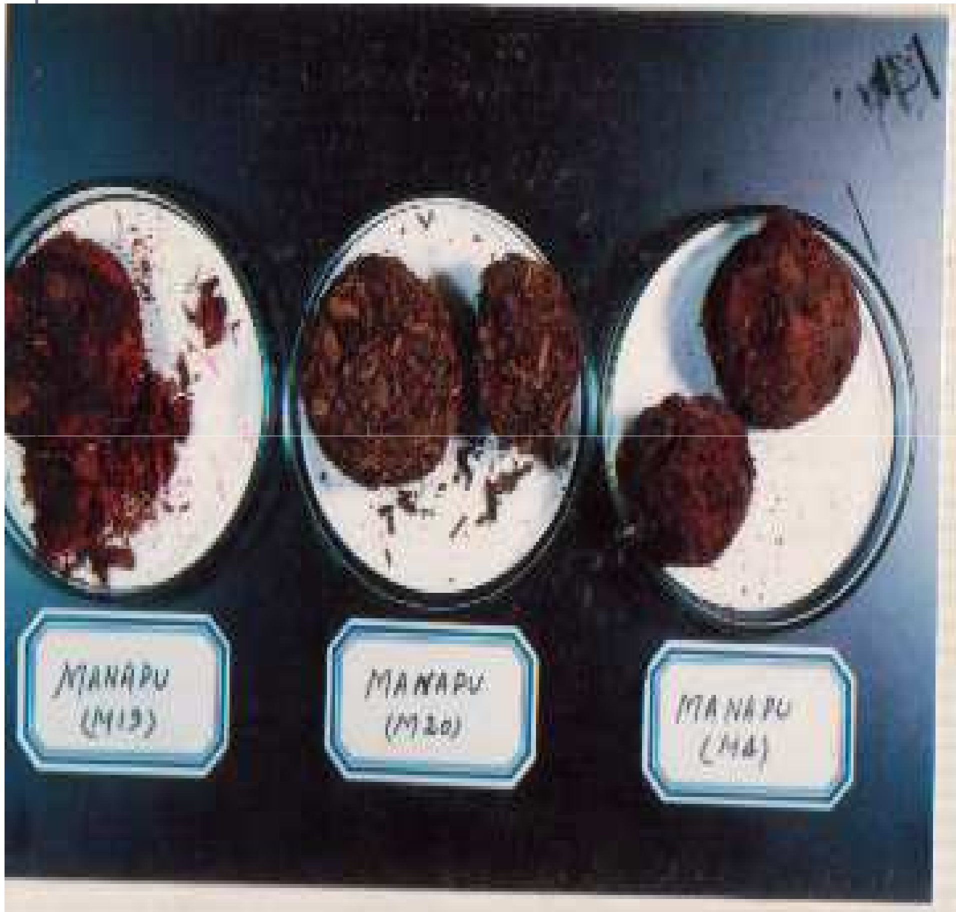

Kneading - II, 20 min
Preparation rounded cake (3-4 cm diameter)
Placing onto straw on the floor
Covering by straw
Fermenting (30-31oC, 5-7 days)
Drying in Sun
Manapu (1-1.2 kg)

Dominant Microorganisms

- Manapu:
  - *Saccharomycopsis fibuliger*
  - *Sacchromyces cerevisiae*
  - Ped. pentosaceus
- Mana:
  - *Aspergillus oryzae*
  - Rhizopus
  - Mucor


MANAPU
(MP13)
MANAPU
(MP6)
MANAPU
(M15)
MANAPU
(M17)
MANAPU
(M18)


MANAPU
MANAPU
(M20)
MANAPU


MANAPU
(MP1)
MANAPU
MANAPU
(MP3)


MANA
(RICE)
MANA
(RICE)
MANA
(RICE)
MANA
(WHEAT)
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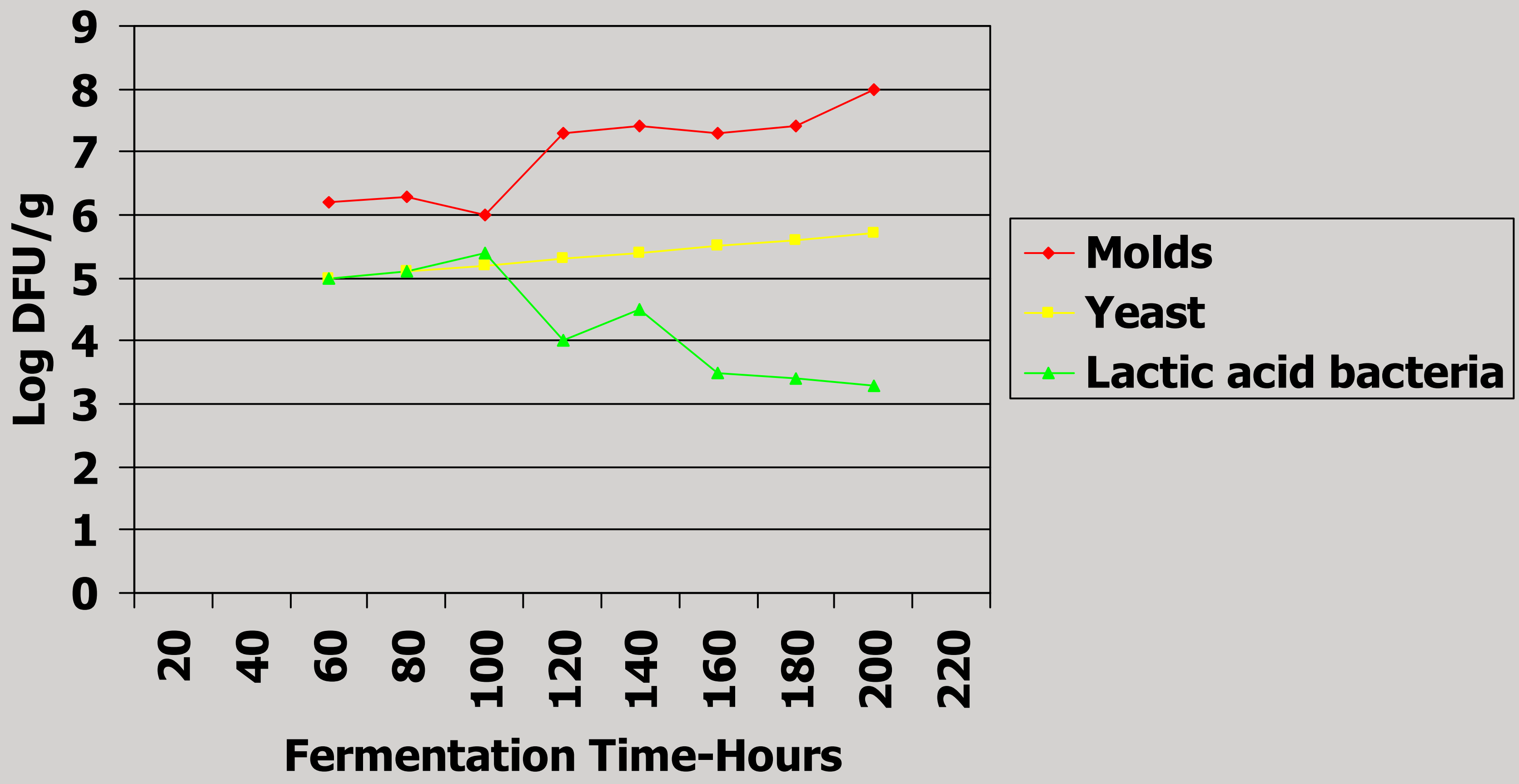
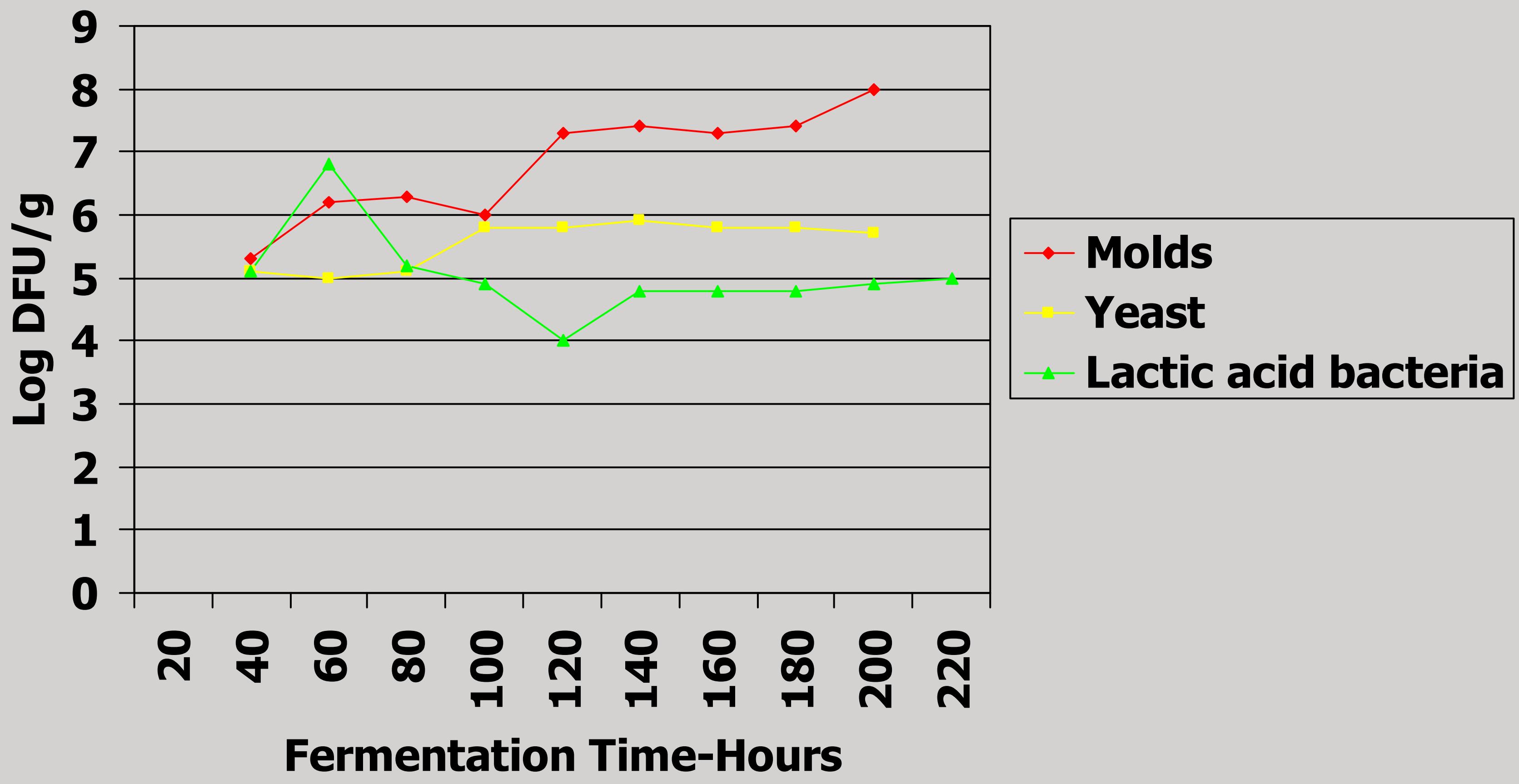
(WHEAT)
MANA
(WHEAT)

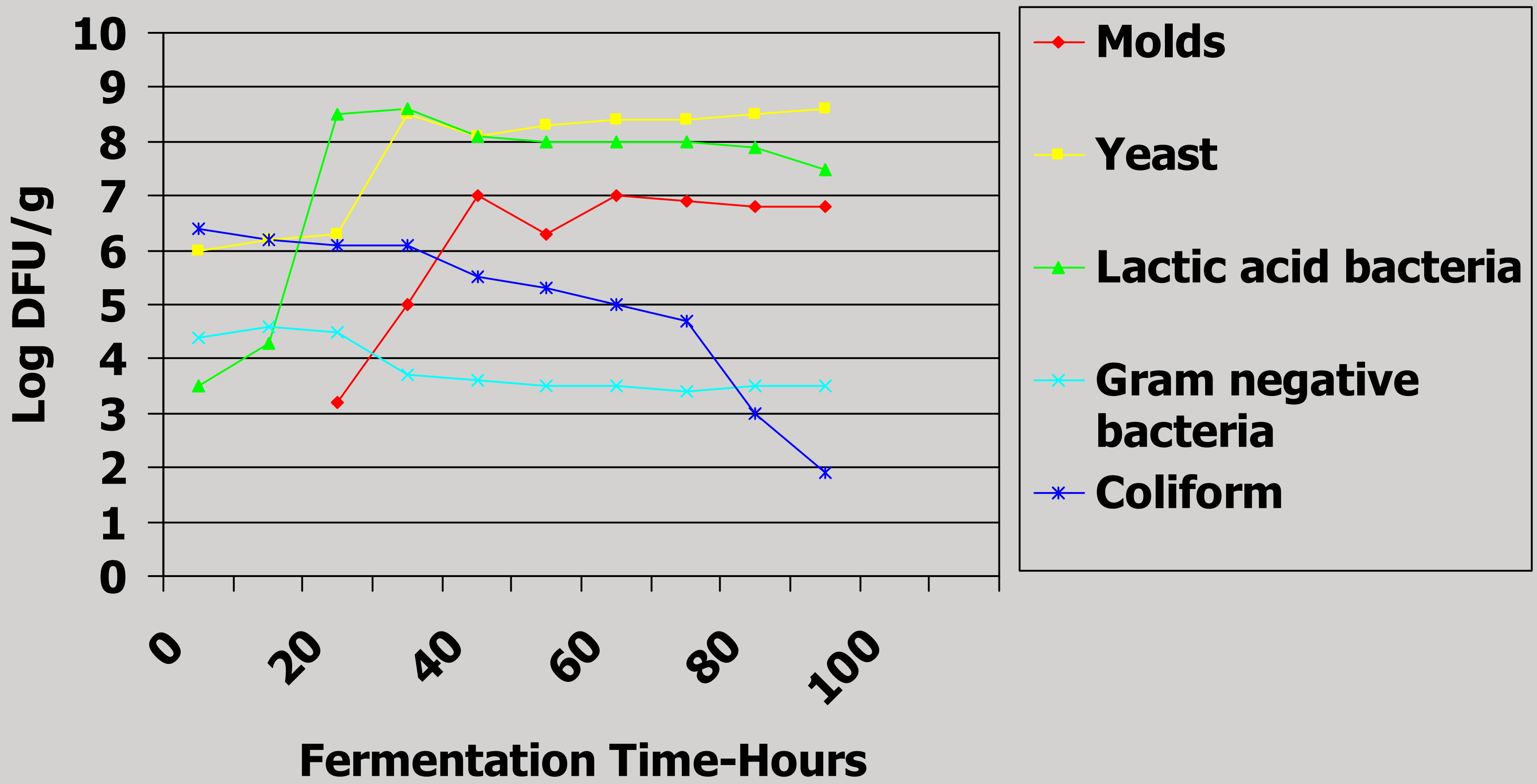
Mycofloral alternations during Murcha (Wheat Mana) fermentation



Mycofloral alternations during Murcha (Rice mana) fermentation



Mycofloral alternations during Murcha (manapu) – Nepalese Yeast cake manufacturing or making



Screening/Selection of best strains from Murcha starters

- Selected best strains of yeast/mold for higher sachharifying activity
- Screened liquifying yeast for alcohol production
- Selected lactic acid bacteria for managing production
- Selected wine yeasts from Murcha are used for wine fermentation and wider production

Development of improved Murcha starters

- Prepared inoculum of selected yeast, mold and lactic acid bacteria at log 6-7
- Prepared Murcha as per traditional procedure
- Tested for the performance of Murcha starters
- Results were promising and developed starter can be prepared

THANK YOU