BC Centre for Disease Control
An agency of the Provincial Health Services Authority

Environmental Health Services

Food Issue

Notes from the Field

Salt koji (shio-koji) fermentation

Request received from:	Vancouver Coastal Health Authority
Date of request:	August 18, 2017
Issue *(brief description)*:	Provide a safety assessment of the recipe and process for an operator wishing to sell a fermented salted rice product, salt koji, in farmers' markets

Summary of search information:

1. Internet sources: google.co.jp proved more helpful than searching North American google sites (translation into English possible). Google scholar.
2. Ovid (define your search terms): not done / time pressure – reviewed existing materials in ENL
3. Review relevant chapters from ethnic food safety on-line text

Background information:

Koji is the result of a fermentation of either soybeans, rice or wheat with a fungus, *Aspergillus.* There are many different types of fungal strains and variants of koji. Koji becomes the precursor to other brine fermented products such as miso, and soy-sauce (shoyu).[1] Differences in the ratios of the types of substrates (rice, wheat, soy) is one of the reasons that Chinese and Japanese soy-sauce has different flavor, colour and aroma.[1] In China, koji is known as *Qu* (麴) and Japanese language uses the same symbol.[2,3] Both types share the taste component of umami, the fifth basic taste (sour, bitter, sweet and salty are the other four).[3] This taste component is in large part attributed to the action of *Aspergillus* which breaks down carbohydrates through the action of amylases and proteases. Carbohydrate substrates such as rice and soy are fermented into more palatable glutamic acid, sugars, and other amino acids.[3]

The overall koji process and subsequent fermentations are well described in the literature, in fact, koji was first chronicled in Asian literature over 2,500 years ago.[1,3] Today, salt koji, also known as shio-koji, is marketed as a healthier type of salt flavouring for foods.[4] Special rice is sold (rice koji) that already has the fermentative enzymes added, and salt koji may be made in the home, or purchased from retailers.[4,5] The name 'koji' is also described by some chefs as the actual *Aspergillus* bacteria. Novel processes using koji by modern chefs include coating meats, fish and other charcuterie products with *Aspergillus* to impart unique flavor and texture profiles which will not be the subject of this food issue note.[6] In this food issue note, koji will be referred to as the overall process as originally described by Fukushima in

1979.[1] In fact, *Aspergillus orzae* koji strains [genus=*Aspergillus,* species=*orzae*, strain=koji] have been selected and specialized based on ability to impart specific sensory profiles to soy sauces.[7]

What are the risks associated with salt koji (shio-koji)?

The process for koji, as described by the operator, involves several steps prior to fungal inoculation and the first fermentation for 48 hours at 30 to 38°C [85 to 100°F]), then addition of salt prior to a secondary fermentation for seven days at room temperature. A diagram of the proposed salt koji process, alongside a defined koji process is shown in Figure 1.

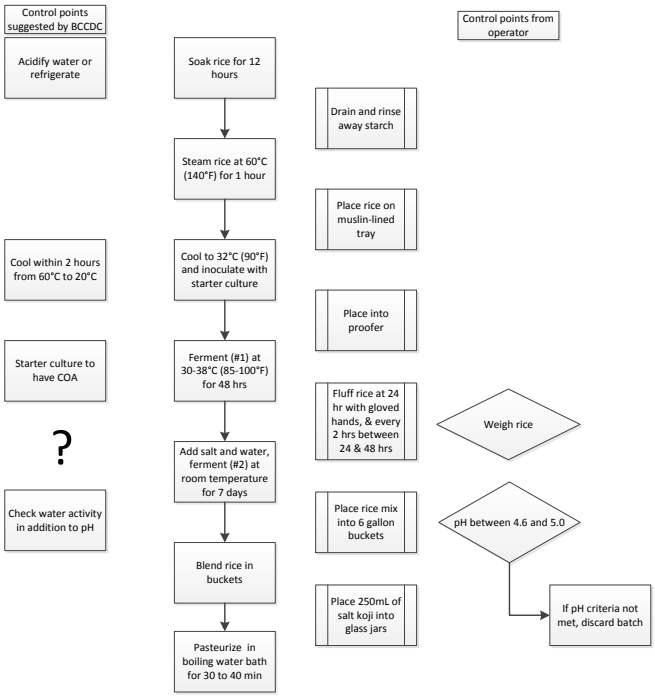

Figure 1a. Food process mapped from food safety plan provided by the operator

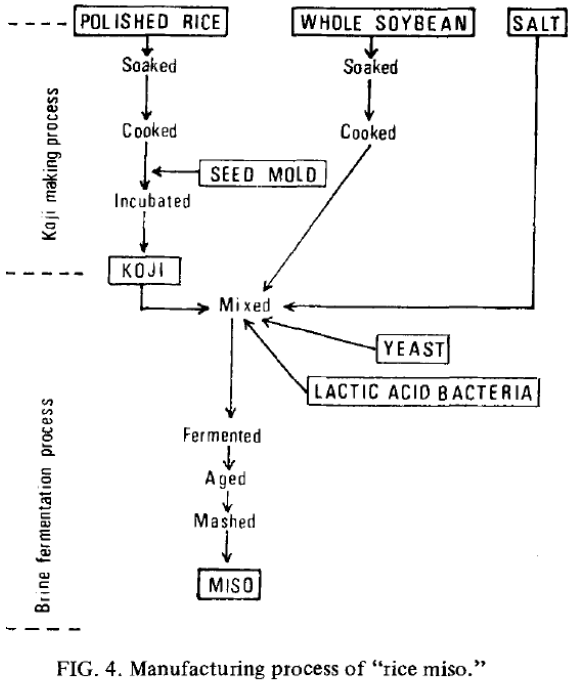

Figure 1b. Figure 4 from Fukushima, 1979[1]

The salt koji process received from the operator was mapped out to define the process steps (Figure 1a). This process appears to follow the process for manufacture of miso paste, before a full fermentation is conducted, and without addition of soybean ingredients, yeast and lactic acid bacteria (Figure 1b, right).

Risks associated with cultured (fermented) koji rice and soy products include *Bacillus cereus,* various *Staphylococcus* spp., *Pantoea* spp., *Enterobacter* spp., mycotoxins and biogenic amines.[2]

Previous guidance on koji or Aspergillus fermentation from British Columbia

While no previous guidance on koji has been given, a recent review of tempe, a soybean product fermented with a different fungi, *Rhizopus*, shares food processing control points.[8] These control points (similar to tempe) are shown in the bullet points below, and described in the left hand side of Fig 1a.

- Soaking of rice (or any grain such as soy or wheat) should be done in a manner that does not amplify any existing hazard. Although the rice will be cooked, spore-forming organisms are a concern, specifically for *Bacillus cereus.* Also, some strains of *B. cereus* have the ability to form a heat-stable emetic toxin (cereulide) that is not inactivated by heat. To control this risk, soaking water should be either acidified to a pH of ≤4.6, or the soaking should occur at refrigerated temperatures of 4°C or less.
- Following cooking and before culture inoculation, the rice should be cooled following normal recommendations (i.e. cooling between 60°C to 20°C occurs in 2 hrs or less). The fermenting agent should be added to the rice in two hours or less once the rice cools to below 60°C. The target temperature for fermentation of this product is 32°C.
- Starter cultures should be defined as to the fungal, bacterial species present, and should only be purchased from companies that can provide a certificate of assurance (COA) that the cultures have been tested free of pathogens. No back-slopping or use of previous cultures are permitted, and new culture used for each new batch.
- The operator has stated that the final pH of the product should be within 4.6 to 5.0, otherwise the batch will be considered to have failed and will be discarded. In addition to pH, we recommended that the water activity be measured, both at the start and end of shelf-life to determine temperature storage conditions suitable for the product. The water activity range supplied by the operator following this request was 0.82 to 0.86 for two batches, one made in the last week and one made the previous year.

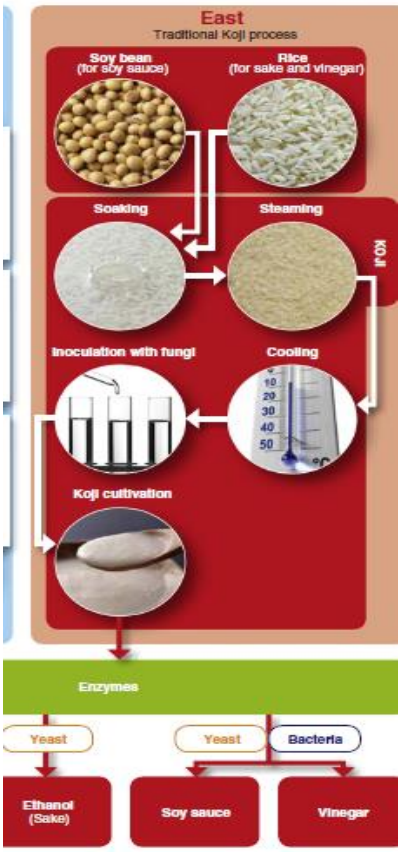

Figure 2. Basic koji process (Zhu and Tramper, 2013)[3]

A question mark is also shown in figure 1a. where further information was requested from the operator. Questions about the proposed process include (1) during the secondary fermentation, is any additional culture added or is the *Aspergillus* fermentation intended to continue? Or, does the original culture mix contain LAB? In the traditional process (shown in 1b.) lactic acid bacteria is added with the addition of salt, along with yeast in the "brining" fermentation step. Any naturally occurring LAB would be destroyed once the rice is cooked, and likely out-competed once the *Aspergillus* culture is added. However, if the original culture mix contained LAB, even small amounts would be sufficient, once salt is added, to allow for secondary fermentation. In response, the operator explained that no LAB was added or present in the starter culture, and the purpose of the salt was to inhibit and stop the *Aspergillus* fermentation during the 6 to 7 days after addition of salt. Active fermentation would be inhibited in one day, and all evidence of fermentation over at the end of one week.

The second question for the operator was a note in the food safety plan that the weight of rice will be measured (why?), but the rationale for this is not given.

The assumption would be a specific amount of salt to rice ratio (w/w) is required: while further review of the literature may offer answers, the operator was asked to provide some guidance to inform this part of the process. In response, the operator informed the inspector that the weight of rice after the first 48 hr was measured so the correct proportion of salt could be added. This control point, and further details on the exact process were requested in the food safety plan. Note that a ratio of 5:1 (w/w) rice to salt would result in 16.7% salt content.

Previous guidance on koji fermentations from reputable literature sources

A recent summary review of the food safety risks involved with fermented soy products (which may not be directly applicable to rice fermentation) include:[2]

1. Contaminated raw materials
2. Lack of pasteurization
3. Poorly controlled natural fermentations
4. Suboptimal fermentation starter cultures
5. Inadequate storage and maturation conditions
6. Consumption without prior cooking

Previous guidance on storage conditions for potentially hazardous foods (PHF)

The operator expects that the final pH of the product will not be below a pH of 4.6, and this is traditionally described as a potentially hazardous food (PHF). A combination of pH, water activity, final salt concentration and pasteurization conditions informs how a product is stored. Once the operator measured the water activity, we were able to define product storage, described below.

According to the FDA food code (2013)[9], foods that are suspected of containing spores that have been heat-treated and packaged can be stored at room temperature if they meet specific pH and water activity requirements (see Figure 3). The operator's water activity for the salt koji, if less than 0.95, would be allowable for storage and sale at room temperature. Following a request to provide results on, two separate batches of the product's water activity and pH met this requirement: water activity (ranging from 0.82 to 0.86), pH (both batches at 5.09). The operator has prepared a label asking for the product to be refrigerated once opened, and this is an acceptable practice.

Table A. Interaction of PH and A_w for control of spores in FOOD heat-treated to destroy vegetative cells and subsequently PACKAGED

A_w values	pH: 4.6 or less	pH: > 4.6 - 5.6	pH: > 5.6
≤0.92	non-TCS FOOD*	non-TCS FOOD	non-TCS FOOD
> 0.92 - 0.95	non-TCS FOOD	non-TCS FOOD	PA**
> 0.95	non-TCS FOOD	PA	PA

* TCS FOOD means TIME/TEMPERATURE CONTROL FOR SAFETY FOOD
** PA means Product Assessment required

Figure 3. FDA Food Code (2013), pg. 22 Time temperature control for safety[9]

Other recommendations for storage of pasteurized and salted foods can be found in the FDA Fish and Fisheries Hazards guidance, although whether they can be applied to salt koji is uncertain. For sauces and surimi type products, refrigerated storage is permissible under the following conditions:[10]

- 5% salt
- A pH of 5 or below
- A_w of 0.97 or below

These conditions, as long as correct pasteurization is achieved, will mitigate the hazard of *Clostridium botulinum*, when the product is stored under refrigeration. Based on results from the operator, salt koji has a salt content of >15%, and A_w <0.9, therefore is acceptable. Although botulism is not listed as a hazard for salt koji, once the jar is opened by the consumer in the home, it is possible it could be contaminated by the consumer. The operators plan to provide instructions to refrigerate are good, and appropriate with similar advice given for these products by commercial suppliers.[4]

Recommendations from BCCDC:

The salt koji process is based on traditional methods that have existed for over two thousand years – however, the operator's process steps, although stated, did not completely describe the purpose of the secondary fermentation. To determine process safety, additional information was requested. In this application, the operator helped speed the approval by supplying information about the overall process. Other controls, as suggested previously include:

1. Starter culture ingredients should come with a COA to demonstrate absence of pathogens, particularly spore-forming organisms such as *B. cereus,* and *C. perfringens.*
2. The soaking water should be either acidified or the rice / water mixture should be refrigerated to prevent growth of pathogens
3. Cooling time should be monitored before the cooked rice is inoculated. Inoculation with fermenting agent must occur within two hours or less once cooked rice is cooled below 60°C to the target fermentation temperature of 32°C.
4. To determine if the pasteurized final product can be stored at room temperature, both pH and A_w tests at a minimum, are required for this product. Salt concentration may also be considered. In this application, although the operator provided a pH result, they were additionally requested to provide water activity test results, at the time of bottling and at the expected end of shelf-life date, before a final assessment of product safety could be determined.
5. The boiling water bath method of sealing the product is acceptable, but will not destroy spores. As this is a reduced oxygen packaging (ROP) method, refrigeration of opened product, as declared on the label is advisable.

Recommendations for BCCDC

1. Further readings are required to understand this process. Challenges with this ethnic food fermentation is that specific control advice for these products do likely exist, but are not explicitly listed in English literature. An OVID search still needs to be done and reviewed.
2. Opinions on this draft from knowledgeable persons are advised. As this koji salt has been approved in US facilities, BCCDC should attempt to reach food safety authorities there for guidance.
3. As previously suggested in the raw foods issue note,[11] compilation of fermentation related assessments and control points would be helpful.

Lessons learned

1. It is often a useful practice to engage with operators who are knowledgeable about their process.
2. When researching an ethnic food, search that countries google search engine – it is more informative as local google search engines are optimized to the location you are searching from.

References

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