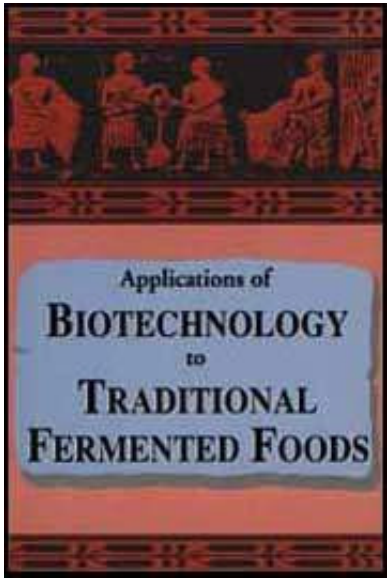

Applications of Biotechnology to Traditional Fermented Foods (BOSTID, 1992, 188 p.)

V. Animal derivatives

17 Using Mixed Starter Cultures for Thai Nham

Pairote Wiriyacharee

Nham is traditionally made from fresh lean pork that is trimmed; minced; mixed thoroughly with salt, potassium nitrate, cooked rice and seasonings; and packed in either banana leaves (1) or cylindrical plastic bags (2). Nham production in Thailand depends on chance contamination with wild organisms - lactic acid bacteria and nitrate reducing bacteria. It is a long process; generally the

fermentation lasts 3 to 5 days depending on the season. When nham is packed into cylindrical plastic bags, which exclude air, and is held in the bags during fermentation, a microenvironment is selected for microorganisms that are not only salt tolerant but can also grow in the absence of air. In these gram-positive fermentative types of microorganisms, lactic acid bacteria are predominant (3,4). The fermentable carbohydrates are used by those organisms to produce organic acids, mainly lactic acid, that contribute to a variety of flavors and textures. The nham finally develops approximately 1.0 percent total acidity as lactic acid and the pH is 4.3 (5).

MARKETING PROBLEMS

Problems in marketing traditional nham include its short shelf life and high price and the intensive labor required for its production. It has high energy costs if kept under refrigeration in the marketplace. Additionally, the manufacturers have a heavy exposure to risk of losing a large stock through a process failure. Pork meat is quite expensive, and the raw material cost is increasing more quickly than the selling price. In addition, large-scale production of nham has the problem of its short storage life. A longer shelf life is required so that the nham can be distributed to the marketplace. Therefore, the nham market needs the product to have consistent quality, safety, and longer shelf life. The nham should stay fresh and not turn rancid or develop an off flavor or change in color when it is in the marketplace.

On the other hand, nham production depends on natural fermentation; the product quality therefore varies from batch to batch. The shelf life of nham is quite short - approximately a week at Thai ambient temperatures. Chilled conditions can extend

the shelf life, but normally the product is stored at ambient temperatures. The sanitation conditions of the processing are also poor because of a lack of knowledge and technology. The initial native lactic acid bacteria may be insufficient to bring about the normal ripening process. This may allow pathogenic bacteria to grow before lactic acid bacteria occur, resulting in the possibility of food poisoning. Since most nham is consumed without further cooking, proper fermentation is of paramount importance in ensuring the product's safety.

Somathiti (6) found that the initial coliform count was high in nham - approximately 107 cells per gram - and decreased to 102 cells per gram on the fifth day. An investigation of Salmonella in nham in the Bangkok market showed that it was present in 56 (or 12 percent) of 450 samples. In nham produced in Chiang Mai, Chiang Rai, and Ubonratchathani, Salmonella was found in 25 percent, 42 percent, and 11 percent, respectively, of the total samples. However, Shigella sp. was not found in nham bought from any of these markets.

Thus, the nham process needs to be studied to improve product quality, to give a more uniform standard quality, and to develop the technology for applying of the process on an industrial scale before launching extensively in the Thai and export markets.

NHAM DEVELOPMENT

In developing of an improved nham process, not only is there a need for the knowledge of modern scientific discoveries and technological developments but also the knowledge of consumers' needs and wishes. The final product must be acceptable to consumers. A unified system is required that combines scientific and

consumer information for systematic development of the nham product.

Effect of Starter Cultures

In our research mixed starter cultures and the carbon sources used in nham formulation were important factors in determining product quality (7). The starter cultures had a potential to make a good nham quality. Cooked rice, a carbon source for lactic acid production by starter cultures, was an important factor in nham fermentation.

The addition of L. plantarum to the nham mass accelerated very distinctly the decrease in the pH of nham. Consequently, the firmness and color developed, influenced directly by acid production. Those findings were in agreement with the work of many researchers (8-12). P. cerevisiae increased the firmness later during the last period of fermentation. The optimum growth of P. cerevisiae is at pH 5.0 (13), the conditions during this period allow good growth and acid production causing the increase in firmness. L. plantarum inoculation had a very distinct effect in terms of firmness development when it was used together with P. cerevisiae.

M. varians in the nham system significantly reduced nitrate to nitrite during the initial fermentation and increased the tristimulus values at the beginning of fermentation. L. plantarum then continued to intensify the color. This finding agreed with the work of Deibel et al. (14); they reported that nitrate-reducing activity generally occurred during the first 2 to 16 hours, while acid production was initiated after 8 to 16 hours. It was clear that it was important to ensure the nitrate reducing activity of the M. varians that took place prior to its inhibition by

the growth of lactic acid bacteria. The nitrite formed was decomposed spontaneously in acid surroundings into nitric oxide, which subsequently reacted with myoglobin to form a pink compound - nitrosomyoglobin. So the residual nitrate in the nham system reduced quickly when acid was produced. The rate of nitrosomyoglobin formation increased with falling pH, and this reaction takes place best in the pH range of 5.0 to 5.5 (15) and was therefore accelerated by L. plantarum. The L. plantarum inoculation had a very distinct effect in terms of color development when it was used together with M. varians.

On the other hand, L. brevis seemed to be a poor lactic acid producer and decreased the color of the product and also produced gas, which decreased the firmness of the nham.

Microbiological Quality

The starter cultures L. plantarum and P. cerevisiae increased during the initial fermentation and were highest on the third day of fermentation with 106 to 109 cfu/g-' (colony forming units) and then decreased slowly during the later period of fermentation. In the nham sample, on the other hand, the M. varians decreased during the fermentation approximately 2 log cycles by the third day. The total bacterial count was related to the starter cultures counts, but there was a little higher count of approximately 1 log cycle. No yeasts or molds were detected in the finished nham.

The pathogenic bacteria, including Enterobacteriaceae and Staphylococcus aureus, decreased during fermentation. In the nham fermented for 3 days the Enterobacteriaceae and 5. aureus counts were 102 and 103 cfu/g-', respectively.

FERMENTATION DEVELOPMENT

In Thailand large amounts of cooked rice are added to the raw nham mixture. It is degraded only slowly and may result in growth of undesirable organisms during fermentation, particularly at high ripening temperatures. Glucose is therefore added to the cooked rice. This ensures a sufficiently rapid initial growth and nitrate reduction by M. varians and rapid later pH drop, without inhibiting the chemical reactions necessary for the development of firmness and desired color.

Cooked rice and glucose had no effect on pH reduction during nham fermentation. As the fermentation time increased, the pH decreased. The pH dropped rapidly after 18 hours of fermentation at 30°C, 43 percent relative humidity with pH 5.1. The beginning of cooked rice reduction coincided with the increase in reducing sugars after 12 hours of fermentation. The reducing sugars declined after another 12 hours of fermentation, and this coincided with the decrease in pH. This indicated that if both cooked rice and glucose were used at high levels (10 percent and I percent, respectively) at the beginning of the fermentation, the pH dropped more slowly than if lower levels were used (8 percent cooked rice and 0.5 percent glucose). Increasing the amount of cooked rice, on the other hand, reduced the firmness of the nham. There was an increase in weight loss at the high level of glucose. There were 1.0 to 1.3 percent reducing sugars and 2 to 3 percent cooked rice in the finished nham, and this residual carbohydrate could be used by the undesirable organisms during storage. Therefore, the carbon source levels in nham should be reduced.

When the glucose level was maintained at 0.5 percent but the level of cooked rice increased, a longer period was required to attain adequate fermentation end

products (16).

It was also found that 6 percent cooked rice with 0.5 percent glucose in the nham formulation, when fermented with starter cultures at 30°C and 97 percent relative humidity, caused rapid pH reduction. Acid production was good, firmness and color development were satisfactory, and the product was microbiologically safe.

The rate of fermentation and the ultimate pH of nham are directly influenced not only by the specific formulation but also by the processing conditions. Since the safety and quality of nham depend on the rate and extent of acid production, a thorough understanding of these environmental parameters is essential for total control of the product. In our research, higher temperatures increased the rate of fermentation, reduced pH, and improved firmness and color development. The initial temperature of nham was very important in determining the final product. The achievement of lowering pH was affected by the initial product temperature and the time at that temperature. For experimentation with frozen meat, the temperature of nham mixtures was 15°C; with fresh meat the temperature was 26°C. The pH dropped more quickly in nham made with fresh meat than with frozen meat.

Nham made using frozen meat was fermented at 30°C and 97 percent relative humidity. It took 3 days to reduce the pH to 4.3 to 4.4, while the nham using fresh meat fermented under the same conditions needed only 2 days to reduce the pH to 4.1.

Nham is usually held at a high temperature during processing to ensure rapid fermentation, but this can also accentuate the growth of pathogens. In addition,

nham is usually eaten without further cooking by the consumer. These conditions make strict control of the product essential. Although proper sanitation, employee hygiene, and the control of raw materials definitely reduce contamination, ultimate control of product safety must be inherent in the formulation and process. The addition of starter cultures can provide sufficient microbial numbers to ensure numerical dominance over the natural flora, including pathogens, and in combination with the proper processing controls can guarantee the safety and quality of the final nham.

Shelf Storage

Nham is usually sold in Thai markets at ambient temperatures (20° to 30°C). It was found that nham prepared using the improved conditions described here when stored at these temperatures had a shelf life of 9 to 11 days while commercial nham usually has a shelf life of only 3 days. In supermarkets nham is stored at chilled temperatures (5°C), and it can be exported at low temperatures (1°C). Additionally, consumers usually store the product in a household refrigerator (10°C). It was found that shelf life was extended to 63 to 103 days at storage temperatures of 1° to 10°C. The higher the storage temperature, the greater the change in nham quality.

Sensory Evaluation

Nham fermented with 103 cfu/g M. varians, 103 cfu/g L. plantarum, and 106 cfu/g P. cerevisiae with 6 percent cooked rice and 0.5 percent glucose at 30°C, 97 percent relative humidity for 3 days, was accepted by the trained panel, with an overall acceptability mean ideal ratio score of 0.95+0.01. For quality degradation

during storage, the overall acceptability of the product depended on sourness and off-flavor detected in the sample.

Nham using fresh meat fermented at a low temperature was given a higher than ideal score for sourness. However, the newly developed formulation for nham was superior to that of the commercial nham.

The consumer panel was also used to determine the effect of reducing the fermentation time from 3 days to 2 days. The results showed that only visual texture was significantly different from the ideal product.

In consumer testing the majority of the consumers (90 percent) accepted the developed nham in terms of sourness, spiciness, and saltiness.

In conclusion, the development of traditional fermented pork sausage, nham, was very successful in that the product was developed by using mixed starter cultures and had a very high quality in terms of consistency, microbiological safety, and longer shelf life. It was also acceptable by the target consumers. The product could be processed in a simple plant and with equipment that was available at the fermented meat factory with only an improvement in the technology of culture preparation and temperature control. In addition, the developed nham had a longer shelf life than commercial nham. The product, therefore, could be shipped from the cottage industry producers in the north to all provinces in Thailand, particularly to Bangkok, and also gave the potential for overseas shipment if refrigeration is used.

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18 Starter Cultures in Traditional Fermented Meats

Margy Woodburn

Fermentation traditionally offers an easy and low-energy preservation method for meats that results in distinctive products that have an important part in the diet of people making them. Such fermented meats contribute both nutritional value and pleasure to meals. However, products are not the same from time to time. Indeed, the product may spoil, cause illness due to pathogenic microorganisms or their toxins, and even become lethal due to botulinum toxin production if the normal beneficial microbial flora do not multiply as usual. To prevent these problems, the use of starter cultures has become commonplace in many countries, including developing countries.

One example of such fermented meat is nham, a traditional Thai sausage. Nham is made by mixing salt (3 percent by weight) and garlic with ground lean pork. Nitrate and nitrite salts also are added in commercial production. The mixture is then wrapped in a banana leaf or stuffed in cellulose tubing. Fermentation is at ambient temperature (about 30°C in Thailand) for 3 to 4 days, after which it remains in good condition for only 1 to 2 days without refrigeration. Since nham is frequently eaten raw, it is important that pathogenic bacteria be killed as well as that botulinum toxin and staphylococcal enterotoxins are not produced. Since hogs are frequently infested with Trichinella spiralis, these larvae should not be viable.

A study was conducted on nham made with and without the addition of one of two levels of a commercially available dry starter culture preparation (Griffith Laboratories, Ltd., Thailand) (1). Portions in polyethylene film bags were inoculated, sealed, and incubated at 30°C. The inoculum was S. aureus (a mixture

of three enterotoxin-producing strains) and E. cold (three strains). Microbial numbers, pH, and titrable acidity were determined at intervals during the fermentation. The meat used was from two hogs that had been experimentally infected with trichinae at weaning; viable trichinae were determined at 24-hour intervals.

S. aureus was able to multiply (10x) and remain viable only in the control inoculated samples. E. cold was not detected at 96 hours in the sausage made with the higher level of starter (1.5 percent by weight) and had decreased greatly in products made with the 0.75 percent level. The use of the higher level of starter preparation resulted in loss of infectivity of the trichinae larvae, although further research is necessary to confirm this effect. The addition of starter culture resulted in more rapid acid production and slightly lower end-point pH.

It is important to keep in mind that natural fermentations are difficult to replicate in other settings. For example, the meat mixture for nham is traditionally wrapped in small banana leaf packets. The leaves contribute to the surface flora of the sausage, which no doubt changes the fermentation pattern. Flora of work surfaces and of the pork itself may be different.

Drying often follows fermentation of similar meat products to provide for long-term preservation. Dendeng ailing, Indonesian seasoned beef that has only a traditional short fermentation period before drying, was found to have a lower pH and total gram-negative bacteria, staphylococci, and E. cold counts when prepared with a starter culture of Lactobacillus plantarum than in the traditional manner. Those with a starter culture dried more rapidly at 50°C and had lower water activities (2).

The effectiveness of lactic acid bacteria in suppressing the multiplication of undesirable microorganisms is largely attributed to the production of organic acid. However, additional factors include the production of bacteriocins and hydrogen peroxide. More general effects include competition for essential nutrients.

To maximize the quality, reproducibility, and safety of the product, strains of bacteria are selected based largely on the qualities of self stability and viability as used, rapid acid production, and desirable product qualities. As in the starter culture preparation used for nham, strains of Lactobacillus and Pedicoccus are the most common (3,4). The compatibility of strains is important, which includes resistance to or lack of production of bacteriocins. In addition to tolerance to the salt and nitrite levels of the mixture, the culture must be active in the temperature range used for the fermentation. The product must have the expected palatability characteristics. No harmful compounds may be produced. These same attributes can be more efficiently arrived at through the application of the techniques of molecular biology.

The success of traditional fermentations depends on the complex interaction of the food components, the natural flora of the ingredients, and the surfaces in contact with the food, atmosphere, and ambient temperature. Our knowledge of these conditions is still limited for many of the fermented meats. Alaskan outbreaks of botulism from native sea and land mammal products may have increased as plastic bags became the common container and the fermentation rate was speeded by placing the container near the stove (5). Thus, as transitions occur from traditional fermentations to new adaptations, knowledge of the basic processes becomes essential.

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19 Fermented Fish Products in the Philippines

Minerva SD. Olympia

In many parts of the world especially in Asia, fermented foods are popular and well liked by the general populace and so widely used that the daily diet of the people would not be complete without them. In a developing country like the

Philippines, where many fermented food products are known, their popularity is due not only to their characteristic flavor but also to the fact that other processing methods, such as freezing and canning, are generally expensive.

Despite their popularity, research and development on fermented foods is meager. Most of the traditional food fermentation industries especially in the Philippines are rural, seasonal, labor intensive, informal, and capital deficient. Commonly, fermented foods are sold and consumed in the areas where they are produced.

The methods of processing were developed in homes and improvements were based on the observations of the practitioners. Fermentation processes are normally handed down from generation to generation. There is little interest in knowing the role of microorganisms and the physical and chemical changes that occur in the products. What is recognized are changes in color, odor, and taste that result from modifications of the process or variations in the ingredients or conditions. Most processes are conducted on a trial-and-error basis with little quality control. Product quality primarily depends on the experience of the processor.

In the Philippines, fermented fishery products can be divided into two groups. The first group includes those containing high concentrations of salt - about 15 to 20 percent in the final product. This group consists of bagoong (fish paste) and patis (fish sauce). These products are generally used as condiments.

The second group includes burong isda (fermented rice fish mixture) and burong hipon, also known as balao balao (fermented shrimp rice mixture). These products, when fermented, become acidic with a cheese-like aroma.

FISH PASTE (BAGOONG)

Product

Bagoong is the undigested residue of partially hydrolyzed fish or shrimp. It has a salty and slightly cheese-like odor (Figure 1). The characteristics of this product vary depending on the region where it is made and consumed. In the Tagalog provinces, the fish paste is completely fermented and ground, with or without coloring matter added. In the Ilocos region and Pangasinan provinces, the products are either partially or completely fermented. In the Visayas and Mindanao, the product is slightly fermented without liquid; the fish is hard and solid salt is present (1).

Preparation

The fish used for bagoong include anchovies, sardines, herring, silverside, shrimp, slipmouth, freshwater porgy, oysters, clams, and other shellfish. The fish are washed thoroughly and drained well. Salt is mixed with the drained samples at varying proportions from 1:3 to 2:7 depending on the bulk of the preparation. The mixture is allowed to ferment for several months or longer until it develops the characteristic flavor and aroma of bagoong.

Bagoong is eaten raw or cooked and is generally used as flavoring or condiment in many traditional recipes. As an appetizer it is sauteed with onions and garlic and served with tomatoes or green mangoes. In rural areas, bagoong is eaten with vegetables, and, especially in the coastal regions, it is often the main source of protein in the diet.

Microbiological Analysis

Results of earlier studies on the microbiological changes in bagoong showed that the total viable count decreased with time. Aerobic organisms predominate at the onset of the fermentation followed by the microaerophilic and anaerobic microorganisms at the later stages (2,3). Information gathered on the microflora indicated that both desirable and hazardous microorganisms are present in this product.

FISH SAUCE (PATIS)

Product

In the Philippines the production of fish sauce is always accompanied by the equally important product bagoong. This product is the clear supernatant yellow-brown liquid obtained by decanting and/or pressing or centrifuging bagoong after it has been thoroughly fermented. Fish sauce may be obtained either from fish or shrimp bagoong after I to 2 years of fermentation. The longer the digestion period, the better.

Preparation

The raw material used is similar to that of the fish paste. They differ only with respect to the period of fermentation. To obtain the fish sauce, the fermentation is continued until liquid forms on top of the mixture, after which it is drained and filtered.

Microbial Analysis

The total bacterial counts decreased rapidly up to the sixth month and declined slightly until the end of fermentation. Most of the organisms isolated were facultative anaerobes.

Chemical Changes

The solid material is progressively digested with the protein, gradually solubilized by enzyme action, leading to increases in peptides and amino acids in the liquid component. The soluble protein/polypeptide ratio was found to be relatively constant after I month. This suggests that most of the proteolytic activities occurred in the early period. Amino nitrogen and total volatile bases (TVBs) increased steadily until the seventeenth day of fermentation. In addition, the lipids in the fish are believed to be broken down during fermentation to yield fatty acids. These may act as precursors for flavor and aroma compounds and may also participate in the browning reactions that increase with prolonged periods of fermentation.

FERMENTED RICE AND SHRIMP (BALAO BALAO)

Product

Balao balao is a fermented cooked rice and shrimp (Penaeus indicus or Macrobrachium species). The mixture becomes acidic during fermentation, and the shrimp shell reddens and softens. It is commonly prepared for the table in sauteed form and is eaten either as an appetizer or main dish.

Preparation

The general method for making balao balao is by mixing washed shrimp with salt (about 20 percent w/w)and allowing the mixture to stand for 2 hours or overnight. The shrimp are then drained, mixed with cooled cooked rice, and fermented at room temperature for 7 to 10 days.

Microbial Analysis

The total plate count of this product showed a fluctuating trend. It is believed that this is due to sequencing in the flora involved in the process. Changes in the microflora during fermentation overlap, which suggests that there are changes in conditions during the fermentation that lead to the death of one species and the enhancement of others.

Fermented Fish and Rice (Burong Isda)

Product

This product is a popular traditional food in central Luzon. It is usually prepared using freshwater fish. During fermentation the fish flesh becomes very soft and the bones acquire the characteristic softness of cartilage when cooked. Before serving, it is sauteed in oil, garlic, and onion. Similar to balao balao, it is consumed either as an appetizer or as a main dish.

Preparation

The method of preparation is almost identical to that for balao balao. The fish is scaled, eviscerated, and filleted. It is mixed with salt and allowed to stand overnight before mixing with cooled cooked rice. Fermentation is also carried out

for 7 to 10 days at room temperature.

Microbial Analysis

Sequential changes of the bacterial flora also occur in this product and involve the same lactic acid bacterial group as in balao balao.

Chemical Changes

During lactic acid fermentation the major chemical change that occurred was the accumulation of lactic acid from the conversion of carbohydrates. This results in changes in the composition and acidity of the product (4). Such changes are attributed to the lactic acid bacteria, which are also referred to as microaerophiles. Changes caused by microaerophiles do not result in the decomposition of the food to its basic components such as CO2, and H2O (5). Instead, the most common end product of their metabolism is lactic acid.

Research and Development

At present, the technological know-how for the improvement of traditional fermented fishing products in the Philippines is not advanced. This holds true in the case of the burong isda process, which will be described in detail.

Burong isda is a traditional fermented fish product in the Philippines. It is similar to naresushi or funasushi of Japan. Previously consumed as condiment (6), it is now often a main dish because of economic conditions. Burong isda is available in two forms, depending on consumer preferences in a particular area. One is called white burong isda, which has a natural product color, and the other is red burong

isda, which is colored by the addition of angkak or anka. Angkak or anka is a culture of Monascus purpeveus grown on rice. The former is preferred in the western provinces of the central Luzon, whereas the latter is preferred in the eastern provinces.

There are several kinds of burong isda sold in the market, each named for the kind of fish used. One example is burong dalag, a fermented rice-fish mixture using mudfish, Ophicephalus striatus. Other kinds are shown in Table 1. Our particular study deals with burong bangus, a fermented rice-fish product using milkfish, Chanos chanos, or loangus in the vernacular. The method of preparation is shown in Figure 1.

TABLE 1 Lactic Fermented Fish Products in the Philippines; Varieties of Burong Isda

	Name	Local	English	Scientific Name
Burong	ayungin	Ayungin	Silver perch	Therapon plumbeus
Burong	bangus	Bangus	Milkfish chanos	
Burong	dalag	Dalag	Mudfish	Ophicephalus striatus
Burong	gurami	Gurami	Goramy	Osphronemus goramy
Burong	hito	Hito	Catfish	Clarias batrachus
Burong	kanduli	Kanduli	Sea catfish	Arias manillensis
Burong	tilapya	Tilapya	Tilapia	Tilapia nilotica
Balao	balao	Tagunton	Shrimp	Macrobrachium sp.
Burong	hipon	Suwahe	Shrimp	Penaeus indicua

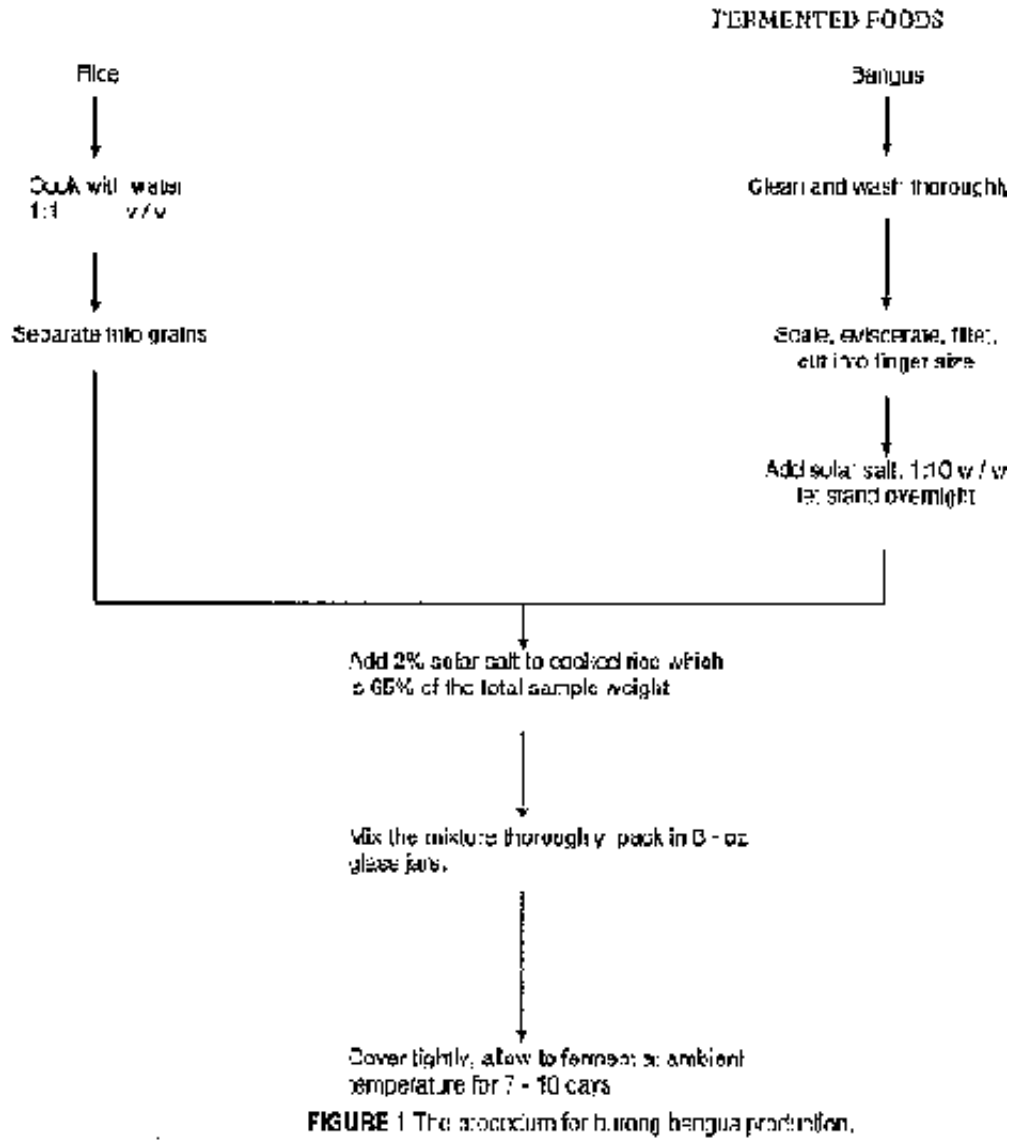

FIGURE 1 The procedure for burong bangus production.

Microbiology

Microbiological analysis of the fermenting mixture showed a sequential type of fermentation with overlapping growth of lactic acid bacteria. The same pattern was also observed by previous workers on other types of burong isda (8-10) and in balao balao (4, 11). This pattern was not, however, observed in market samples. Instead, only one group of lactic acid bacteria predominated in the product from the onset of the process until it was sold in the market. Market samples were also analyzed for the presence of microorganisms of public health significance. Results showed the presence of coliforms Salmonella, and S. aureus.The same results were also obtained by Ferrer (12).

ISOLATING STARCH-HYDROLYZING LACTIC ACID BACTERIA

During our study on the microorganisms involved in the fermentation of burong bangus, some isolates were found to be capable of hydrolyzing starch (10, 13). Relatively few lactic acid bacteria are known to be starch hydrolyzers, and they were mostly isolated from substrates other than fish. In fact, Bergey's Manual of Determinative Bacteriology (14) does not describe lactobacilli as a starch-hydrolyzing bacteria. However, some of the lactic acid bacteria isolated from burong bangus showed otherwise.

The presence of starch-hydrolyzing lactic acid bacteria was also observed in market samples. One of the starch-hydrolyzing isolates, coded L137, was noted to be present in almost all stages of the fermentation process. It was of interest to investigate how the ability of L137 to utilize starch related to its range and level of amylolytic enzyme activity and the type of enzyme(s) that it produces. L137 was characterized and tentatively identified. The enzyme that it produces was also purified. Tentative identification of L137 showed that this strain possesses

characteristics similar to L. plantarum and L. corneyformis subsp. corneyformis. L137, however, differs from these two strains in its ability to utilize starch (15). Earlier studies on the lactic acid fermentation of starch-based products reported that lactic acid bacteria cannot hydrolyze starch. The nonacidformers that predominate in the microflora at the onset of the fermentation process, most of which are amylase producers, first hydrolyze the starch to make it available for lactic acid bacteria (16). However, in some burong bangus samples no nonacidformers were present; yet fermentation went on. The presence of a starch-hydrolyzing lactic acid bacteria in the fermenting rice-fish mixture, especially during the early stages of the process, will ensure a continuous production of metabolizable sugars for subsequent formation of lactic acid. This will result in a rapid decrease of the pH, thereby inhibiting the growth of other microorganisms that may be amylolytic but that might be possible spoilers and/or human health hazards.

Study of the fermenting samples showed appreciable dextrinizing activity. This would indicate that the sugars formed during the process were mostly oligosaccharides. Our study also showed that acidity increases as the fermentation progresses, even with decreasing reducing sugar production. This would mean that the breakdown products of the starch for lactic acid production were oligosaccharides and reducing sugars. The result also indicates that the lactic acid bacteria involved in the fermentation can utilize oligosaccharides to produce lactic acid. Considering the industrial importance of this strain, the enzyme produced by L137 was purified. Results of the study showed that this enzyme indeed produces oligosaccharides when allowed to react with amylose (15). The activity of this enzyme was found to be highly stable at pH 4 to 5 and is optimum at around pH 4.

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20 Fish-Meat Sausage

Sam Angel and Eliana Mora P.

Fermentation allows the preservation of foods of vegetable or animal origin so that they can be stored and shipped at ambient temperatures and used without further preparation. Lowering the pH is the ideal way to process foods for use in less-developed regions of the world. Proteins, which are needed for growth and development, especially in children, are often in short supply in famine-ravished areas and poor underdeveloped countries. In many areas of the world there are frequently underutilized sources of muscle proteins that could provide excellent starting materials for preservation by fermentation or acidulation.

In Germany a popular noncooked fermented sausage (rohwurst) has been produced from beef and pork for many years (1). Lactic acid produced by fermentation lowers the pH of the meat to its gel point which causes it to firm (2). Further drying increases firmness and reduces water activity. The low pH prevents the development of pathogenic bacteria (3,4), and lower water activity prevents microbial growth and spoilage (5).

The pH can also be lowered by using glucono-delta-lactone, which produces gluconic acid upon contact with water, or using citric or lactic acids. Encapsulated acids release acid more slowly and prevent texture breakdown. In the encapsulation process solid acid granules are coated with hydrogenated vegetable

oils or diglycerides, which require heat to release the acid. Graves (6) patented a new water soluble low-temperature release coating for citric acid. The use of acids directly saves fermentation time, and myofibrillar protein gelation can take place within hours after mixing the meat with acids.

RESULTS AND DISCUSSION

Rohwurst beef-pork sausage served as a model for the development of a similar product from underutilized fish, meat trim, and poultry. A sausage-type product allows the combination of muscle from various sources. The object was to use underutilized muscle protein sources, especially fish, to produce a nutritious and acceptable dry sausage. The product was to be eaten out of hand and thus help to alleviate protein deficiency, especially in children.

Underutilized or inexpensive fish, fish tissue residue from filleting operations, red meat trim, and spent layer hens were the raw materials used in Israel, the United States, and Costa Rica to produce fermented dry sausages.

Cod or haddock frames (i.e., skeletons with residual tissue) were mechanically deboned, and the flesh mince was mixed in equal proportions with either beef or pork trim or mince from mechanically deboned spent layer hens. The batters were mixed with salt, sugar, spices, nitrite, and Lactobacillus or Pediococcus starter cultures and stuffed into 20-mm collagen casings. They were fermented at 22°C for up to 24 hours depending on pH development.

Control sausages consisted of beef and pork only. The pH of the fish-meat sausages fell to 5.1 to 5.4, while the pH of the beef-pork controls fell to S.O to 5.1.

Drying took I week, at which time the fishmeat sausages contained 17 to 30 percent moisture and the beef-pork controls 25 to 30 percent. Fat content was 17 percent in all the batters at the outset. After drying it was 21 to 30 percent for the fish-meat sausages and 29 to 30 percent for the beef-pork. The fat contents for the fish-meat sausages were significantly lower than for similar commercial sausages in Germany.

Three panel sessions were held. Between 25 and 70 persons participated in each session. All the sausages were found acceptable, as shown in Figure 1. A minority of the participants commented on a fishy taste, especially for the fish-chicken sausages.

In a 3-year cooperative project (7) the flesh of pond-raised silver carp and sea fish in Israel and Costa Rica was deboned and washed. It was then used to prepare fermented or acidulated dry sausages with pork or beef trim or whole-muscle turkey bottom meat. All-fish sausages and 25 to SO percent fish-meat sausages were prepared. Fermentation was induced with Pediococcus plus Lactobacillus starter cultures. Acidulation was carried out by adding encapsulated lowtemperature-release citric or higher-temperature-release lactic acids.

The pH usually fell to 4.85, except for the fish-turkey sausages where the pH did not fall below 5.0. Starting and final pHs were similar for the fermented and the acidulated sausages, but the pHs for the acidulated sausages fell to their final level within a few hours as compared to several times that for the fermented sausages. Thus, the acidulated blend had a head start on drying. The entire process of pH reduction, firmness development, and subsequent drying was shortened considerably for the acidulated fish-meat sausages.

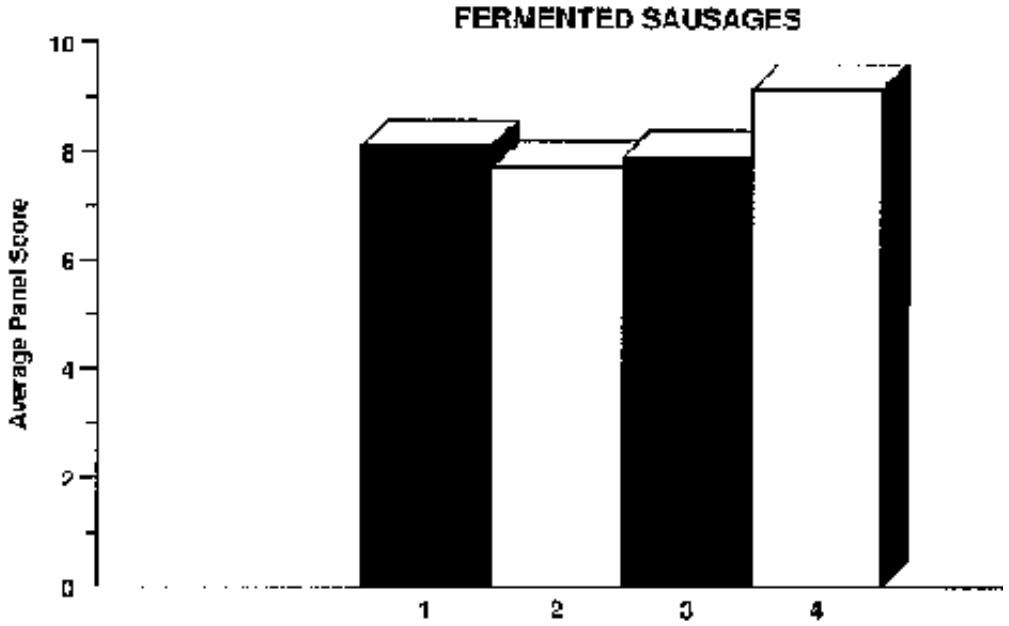

FIGURE 1 Average scores for acceptability of fermented and beef-pork sausages. Average scores of three consumer panels in the United States for fermented sausages made from fish-pork, fish-chicken, fish-beef, and beef-pork. Score of 10 means highly acceptable: 5, not acceptable but not rejected; below 5, various degrees of rejection. Fish-pork sausage received the highest average score, 8.1, of the fish-meat sausages. Next highest average score of 7.9 was for the fish-beef sausages. Fishchicken sausages received a score of 7.7, and nonfish beef-pork sausages received an average score of 9.

Citric acid was found to reduce the pH at lower concentrations than lactic acid, but in 50 percent fish-meat sausages with 0.65 percent citric acid, which was the maximum concentration used, the pH could not be lowered sufficiently. Lactic acid could be used at higher concentrations to lower the pH when necessary. A pH of 4.80 to 4.85 helped the drying process, and lactic acid was of greater benefit in this respect than citric acid.

The experimental sausages were evaluated on a kibbutz in Israel and in 110 households in Costa Rica. The results of the evaluations in both countries were encouraging. In Israel the scores were 5 to 5.6 out of a maximum of 9 for the 50 percent fish sausage. Over 80 percent of the tasters in Costa Rica gave positive responses to the sausages that contained fish (Figure 2). The highest social class was least enthusiastic about the sausages. In Costa Rica the population is not accustomed to eating nonheated sausages. The evaluators therefore either cooked or fried them before eating. The organoleptic tests are consequently being repeated with new instructions.

RECOMMENDATIONS

To minimize production costs, these sausages should contain a minimum of 50 percent fish (from frames or other underutilized sources such as by-catch and trash fish).

Acidulation produced sausage with a good texture, and it can be recommended as a procedure to reduce processing time.

To improve acceptability and nutritional value as well as reduce costs and ensure quality, more research needs to be done on:

· flavor formulation and colorants to meet local population preferences;

· reduction of fat content and introduction of new sources of fat;

· inclusion of soy or other vegetable proteins;

· chemistry and histochemistry of the acidulation and drying processes to improve the efficiency of these steps;

· the effect of replacing nitrite on the wholesomeness of the product [according to Leistner (8), spores of bacilli and clostridia do not grow when there is a sufficiently low pH and low water activity]; and

· protein efficiency feeding for young children and adolescents.

These products should undergo taste tests for acceptability in other Latin American countries as well as other areas with low protein intake.

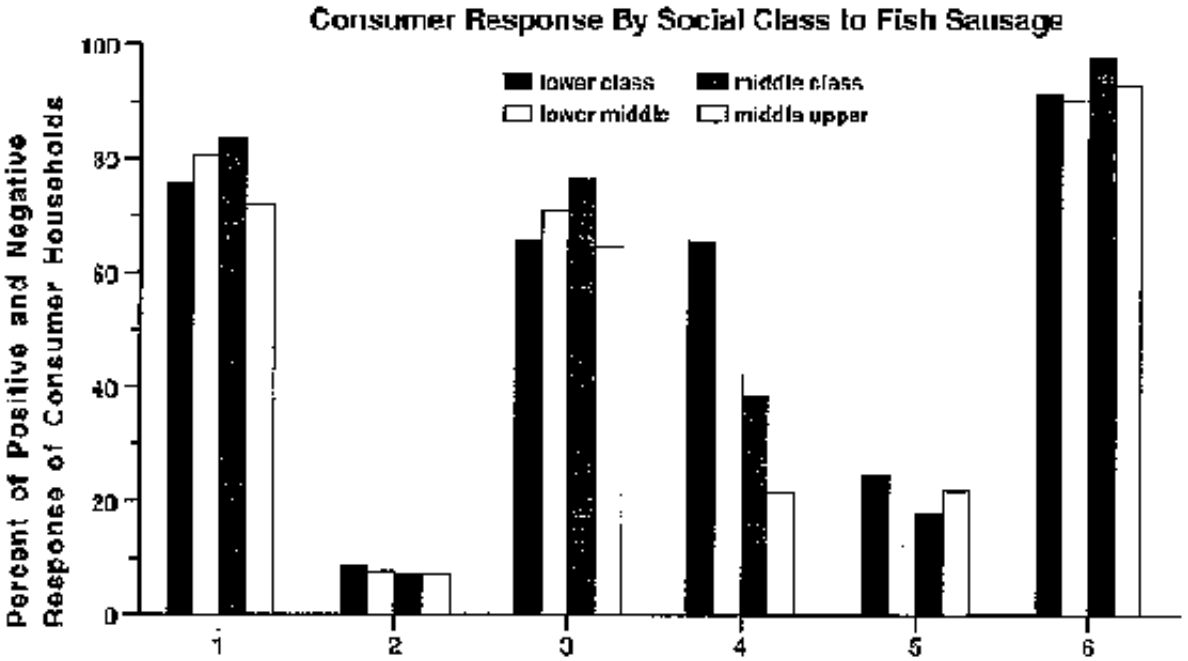

FIGURE 2 Social class of consumers who rated the organoleptic parameters of the fish sausages. Shown are the percent positive and negative and net positive

responses to the fish-containing sausages of over 400 persons in l to households in Costa Rica. Also given are comparisons by social class to other sausages on the market.

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21 An Accelerated Process for Fish Sauce (Patis) Production

R. C. Mabesa, E. V. Carpio, and L. B. Mabesa

The single, probably most important, limitation in the manufacture of fish sauce is the length of time required for its production. It normally takes approximately 12 months from salting to maturity. This limits the turnover rate and overall profitability of a potentially lucrative enterprise. Considering the capital outlay and operating expense required to run a fish sauce business, it is imperative to develop a simple, economical, practicable accelerated process that yields acceptable fish sauce.

With this goal, research and development efforts were undertaken at the food pilot plant of the Institute of Food Science and Technology, University of the Philippines at Los Banos.

OBSERVATIONS

This investigation stemmed from the observation in commercial tanks that freshly drawn fish sauce lacks the desirable aroma of mature sauce; this aroma develops after overnight storage or longer. The appropriate color is there initially but

typical flavor is lacking. It was also observed that flavor, aroma, and color development of palls in both concrete and wooden vats is more rapid and pronounced during the hot summer months. Constant agitation through pumping and frequent transfer of fish sauce from one container to another also hastened and enhanced development of flavor and aroma. It was hypothesized, therefore, that artificial agitation and/or aeration and heat may help with the development of desirable qualities in fish sauce. Thus, small-scale laboratory experiments were carried out initially. It was determined that timing is of primary importance in the application of heat and aeration. The typical fish sauce characteristics did not develop when freshly salted fish was aerated and heated immediately after mixing. Trials were carried out to determine the appropriate time for aeration and heating of the fish salt mixture. It was found that aging for about a month after salting was sufficient and that higher temperatures resulted in more rapid and greater improvement in quality. However, preliminary experiments indicate that the maximum temperature should not exceed 50°C or a cooked flavor results.

A concrete tank simulating the dimensions of a commercial tank was constructed to test the findings in the laboratory. Technical specifications are given below (see box). It was concluded that fish sauce comparable to traditionally manufactured sauce can be obtained in about 2 months or less using modified reaction conditions. These conditions are given under B and C. Sauce characteristics are given under D.

It is likely that production time may be further reduced if strongly halophilic, proteolytic, and thermophilic Bacillus and Pediococcus species used in the laboratory can be used in production.

DISCUSSION

Fish sauce with the desirable qualities of traditionally produced sauce was obtained in the pilot plant. The improved process resulted in an acceptable product in about 2 months instead of the 10 to 12 months required for the traditional process. Clearly, savings in time and an improved turnover rate can result if these results are applied commercially. This means greater income-generating capacity.

Some problems, such as loss of volume and contamination with molds and bacteria, were encountered during heating and aeration. The former was remedied by day-to-day monitoring of fish sauce levels and replenishment with plain tap water when necessary. The second problem was resolved by installing cotton filters at the intake end of the pumps and by adding sorbic acid to the sauce at 0.05 percent prior to bottling.

CONCLUSION

With pilot-level success, there is reason to believe that the process can be applied on a commercial scale. However, there are problems attendant to adaptation of the process. Additional expense will be incurred in equipment acquisition, installation, and operation. Heating and aeration alone will increase the price of palls by about P 50 per drum or about P 0.25 per liter.

TECHNICAL SPECIFICATIONS

A. Tank

1. Type - concrete, cube approximately 0.265 m x 0.265 m x 0.265m I.D.

2. Material - concrete 3:2:1 mixture of sand, gravel, and cement with Sahara water proofing added.

3. Heaters - two 1,000-watt rod-type heaters located close to the center of the tank.

4. Air pump - one aquarium-type air pump with discharge capacity of 5 liters/minute; pump discharge located 2.5 centimeters below heaters.

B. Operating Information

1. Preliminary incubation - 50 days at ambient temperature.

2. Air pump - operated 4 hours a day for 10 days.

3. Heaters - operated 4 hours a day for 10 days.

4. Temperature - 45° to 60°C for 10 days.

5. Power requirement - 7 amperes (pump and heater).

6. Voltage requirement - 220 volts.

C. Raw Material Information

1. Total weight of fish salt mixture - 320 kilograms.

2. Proportion - 1 salt:2 fish by weight (106.6 kilograms salt:213.3 kilograms fish).

3. Fish species - Decapter´'s macrosoma.

4. Source - Navotas Fishery Port.

D. Sauce Characteristics

1. Color - golden yellow-brown highly typical of fish sauce and clear.

2. Odor - slightly acidic and fishy, typical of fish sauce.

3. Flavor - typical fish sauce.

4. Total solids percent.

5. Protein - 14 percent.

6. pH - 6.0.

7. Salt - 24 percent.

8. Specific gravity - 1.21.

9. Yield - 137.5 kilograms.

These costs must be weighed against savings or advantages such as faster turnover rate, decreased overhead, salaries, and power.

Each manufacturer or potential user of a new technology such as this stands to gain substantially despite the additional costs. However, each interested user may find his or her situation unique. A careful study of all terms, factors, and conditions affecting a user should be undertaken before embarking on a new and innovative process such as this.

In light of these results and consequent problems, efforts are under way in the laboratory to reduce process costs, particularly with respect to reducing heating time, minimizing heat losses, increasing heating efficiency, and exploring alternative sources of energy for use in the process.

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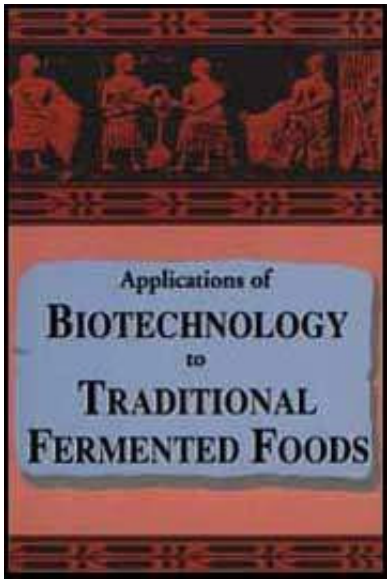

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VI. Human health, safety, and nutrition

22 Nutrition and Safety Considerations

O. Paredes Lopez

Fermentation was one of the first methods used by Man to produce and preserve foods. Microbial fermentations have played an important role in food processing for thousands of years. Fermentations provide a way to preserve food products, to enhance nutritive value, to destroy undesirable factors, to make a safer product, to improve the appearance and taste of some foods, to salvage material otherwise not usable for human consumption, and to reduce the energy required for cooking (1). Preservation of foods by salting is an age-old practice; while preventing the growth of pathogenic microbes, it allows the development of harmless, halotolerant ones that produce desirable censorial changes in the substrate (2).

Traditional fermented foods may be divided into two broad categories: (a) submerged culture-fermentations (SCFs) and (b) solid substrate fermentations (SSFs). In SCFs microbial activity occurs at a relatively low biomass concentration in the liquid phase, while in SCFs microbial growth and product formation occur on the surfaces of solid substrates (3,4). Some examples of traditional fermented foods for SCFs are pulque and tesguino, soy sauce, fish sauce, kaffir beer, and palm and rice wines. Examples of SSFs are tempe, miso, pozol, oncom, and natto. One of the major characteristics that distinguishes SSFs from SCFs is that SSF processes usually occur at low-moisture contents (e.g., 10 to 20 percent),

conditions under which water activity favors the development of filamentous fungi. However, for many indigenous fermentations the microbial interactions are complex and mixed fungal-bacterial, fungal-yeast, and yeast-bacterial combinations occur (5). These interactions play an important role in the nutritional, safety, and sensory characteristics of the end product (6).

EFFECT OF FERMENTATION ON NUTRITIONAL COMPOSITION

Changes in Proximate Composition and Soluble Components

During fermentation the microorganisms secrete hydrolytic enzymes into the substrate and assimilate some of the fatty acids, amino acids, and simple sugars thus liberated. These are converted into microbial structural components and secondary metabolites. Lactic acid fermentation is an ancient process whereby a varied group of bacteria ferment carbohydrates, producing lactic acid as the major end product. This type of fermentation is used for the production of dairy products, sauerkraut, bread, meat, and silage. In particular, traditional SSFs of legumes, cereals, and starchy substrates have been associated in many regions of the world with the activity of lactic acid bacteria (7); during fermentation lactic acid accumulates, with a concomitant increase in acidity and a decrease of dry matter yields. The higher pH values of fermented legumes, compared to other materials under similar conditions, have been attributed to their higher protein content (8,9).

It seems that the only fermented food showing significant changes in its crude composition is pozol. The fermentation mixture contains Agrobacterium azotophilum, which is capable of fixing nitrogen (10). Due to the crude methods of

analysis, the proximate composition of foods does not change much during fermentation. However, there is almost always a high increase in the soluble fraction of a food during fermentation. The proteolytic activity of bacteria in traditional fermentations degrades complex proteins into simpler proteins, peptides, and amino acids. The bacteria used in natto fermentation cause substantial increases in the level of free amino acids and soluble carbohydrates. On the other hand, Rhizopus spp., used in the fermentation of various types of tempe, are highly hydrolytic, and outstanding increases in soluble fat, protein, and carbohydrate are observed. Free fatty acids, including the essential fatty acids, linoleic and linolenic acids, may increase in these indigenous fermented foods (11,12); this increase is thought to be of nutritional significance.

The increase in soluble solids is a nutritionally desirable event, as the food is effectively digested prior to consumption. In some cases the microorganisms are capable of producing pectinases and cellulases, softening the texture of the food and liberating sugars that would otherwise be unavailable to the human digestive system. Consequently, fermented foods are expected to be more digestible than their unfermented counterparts.

Changes in Composition of Amino Acids and Vitamins

Methionine, the limiting amino acid in legumes, has been reported to increase during tempe kedele production, and lysine, the limiting amino acid in cereals, increases during fermentation with Rhizopus spp. (1). During kocho production, an acidic fermentation, the essential amino acid content is considerably enhanced. On the other hand, during tape' ketan and enjera production, the levels of some essential amino acids fall, whereas others remain unchanged (11). In general,

most traditional fermented foods exhibit slight changes in essential amino acids.

Interestingly, isolation of improved strains of Aspergillus niger for an SSF process allowed 200 to 300 percent lysine overproduction compared to the parent strain (5). However, it should be emphasized that bioavailability and balance of amino acids are more important than their total content. Hence, biological experiments to assess their nutritional value are warranted.

Traditional fermentations dramatically improve the vitamin content of a wide variety of substrates. Of all the foods investigated, only enjera showed a decline in vitamin content (1,13).

Changes in Unwanted Components

Unwanted components, such as physic acid, trypsin inhibitor, flatus factors, and lectins, may be present in high concentrations in several desirable foods. Phytic acid and trypsin inhibitor interfere with digestion by binding enzymes. Phytic acid may also bind minerals, reducing their bioavailability. Lectins are capable of binding to the intestinal wall and thus interfering with nutrient absorption. Presoaking and cooking of foods can reduce the levels of some, but not all, of these antinutritional factors. However, microorganisms have the capacity to hydrolyze them, reducing their levels even further (14). Hence, bacteria, yeasts, and fungi that degrade antinutrients at a fast rate and at early stages of fermentation need to be identified or developed (1).

Changes in Biological Value

Since fermentation increases the quantity of soluble proteins in foods, it may

improve the amino acid profile, and because it reduces the levels of certain antinutritional factors that interfere with digestion, it would not be unreasonable to suggest that fermented foods will be more efficiently utilized by the human digestive system. Single- as well as mixed-culture fermentations of pearl millet by yeasts improve starch and protein digestibility (15). Enjera is one of the few traditional fermented foods that shows a decline in protein efficiency ratio (PER), probably due to a decline in the essential amino acid content (16). Also, increases in PER values of some indigenous fermented foods can be obtained by incorporating soybeans into cereal-based substrates.

SAFETY ASPECTS OF TRADITIONAL FERMENTED FOODS

Because many fermented foods are produced using fungi, the risk of mycotoxin contamination is high. During natural fermentations, food-poisoning flora and coliforms may also grow with the lactics. These microorganisms need to be eliminated to make fermented foods safe for consumption (16). Several factors contribute to the safety of fermented foods: (a) Soaking and cooking. Washing, soaking, and cooking treatments reduce the in situ microbial contaminants. (b) Salting. Various fermented foods are made with the addition of salt, which acts as a preservative. (c) Acid formation. Many indigenous fermentations are carried out by acid-producing microorganisms, where these organic acids (e.g., lactic, acetic, fumaric acids) act as preservatives or as bacteriostatic agents. An inhibitory pH for bacterial growth is considered to be 3.6 to 4.1. (d) Antibiotic production. Molds used in some traditional fermentations produce antimicrobial glycopeptides. (e) Low moisture content. In the case of SSF processes, the low water activity may be an important preservative factor. and (f) Reduction of aflatoxin by some microorganisms. Rhizopus and Neurospora species, among others, are reported to

decrease aflatoxin content of contaminated substrates.

Despite these factors, it has been reported that the sanitary quality of some Oriental fermented foods is poor (17,18). Safe products are usually obtained when the following recommendations are observed: (a) appropriate soaking of the beans in acid at a low pH; (b) adequate cooking time; (c) using hygienic conditions during production, handling, and storage; and (d) good refrigeration of products (5°C) between production and consumption.

In summary, production of foods with high nutritional and sensory values, and free of microbiological health risks, is a key component of any policy aimed at upgrading the social role of traditional fermented foods in less developed countries.

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23 Mycotoxic Flora of Some Indigenous Fermented Foods

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Fermented foods have a wide usage in Sierra Leone as baby/weaning foods. Ogi(fermented maize/sorghum) and foofoo pap (fermented cassava) are examples. Foofoo is also one of the two staples of the Creoles that is now widely used by other tribes especially when rice is scarce. Ogiri (fermented sesame seeds) is a favorite condiment used mostly by the poor as a low-cost protein substitute. Several studies (1-4) have shown that toxigenic fungi do not participate in the fermentation processes but contaminate the product during or after the fermentation.

It has been demonstrated (1-4) that at times the substrate for fermentation (maize, sesame seeds) has had prior exposure to mycotoxin. In the case of maize, an aflatoxin B1 level of 200 ug/kg was reduced to 58 ug/kg in the resulting fermented mashogi (5). The long cooking period (6 hours) of sesame seeds before fermentation accounts for the loss of mycotoxins. Studies carried out by Ogunsanwo et al. (6) have shown that losses of 64 percent aflatoxin B. and 83 percent aflatoxin G1 could be observed in ogiri product prepared from Aspergillus flavus-contaminated melon seeds.

In Sierra Leone, ogiri is produced by moist solid fermentation of sesame seeds, a process similar to Nigerian ogiri, which is made from fermented melon seeds (Citrullus vulgaris) (7) and Dawa-dawa from fermented locust beans (Parlkia filicoidea) (8). Traditionally, the boiled seeds are wrapped in jute bags and allowed to ferment for 4 to 5 days before smoke treatment is applied. In such

processes whitish threads are observed after day 2 and molds become obvious after 3 to 6 days (3).

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Microscopic examination of these whitish threads revealed the presence of toxigenic and nontoxigenic Aspergilli and Perticillia species. Detection of the corresponding mycotoxins of these toxigenic fungi in the fermented, marketed, and stored ogiri(4) led to the present study to design appropriate fermentation and storage techniques to reduce the risk of mycotoxin contamination.

MATERIALS AND METHODS

Fermentation Process

Sesame seeds were soaked overnight and pounded in a mortar to dehull. The seeds were then washed and boiled for 6 hours. The boiled seeds were divided into three portions. One portion was transferred to a clean dry nylon fiber bag; the other was placed in a clean dry jute bag. Both were tightly wrapped. The third was placed in a plastic bowl with a tight-sealed lid. Three replicates of each of the nylon fiber and jute bag arrays were made. These were divided into three groups. Group one was left to ferment for 5 days without smoke treatment. Group two received early smoke treatment, from day 2 until day 5. Group three was smoked consistently from day 3 to day 8, and thereafter on alternate days for 2 weeks.

Marketing and Storage

The three common methods for wrapping ogiri are (a) the use of dried banana leaves Musa sapientum, (b) the use of fresh or smoked leaves of the plant Newbouldia laevis, and (c) the use of small plastic wraps.

Leaf and plastic-wrapped ogiri samples bought from the local markets were examined immediately under a stereo microscope. Samples with no obvious fungal presence were selected. Three experimental designs were set up as follows: (a) a set of six samples (three from each type of leaf wrap) was smoked consistently for a week, (b) another set of six (two from each type of leaf and plastic wrap) remained unsmoked and stored at room temperature, and (c) the three types of wraps (minus ogiri) were placed in sterile plastic petri dishes and stored at room temperature.

Determination of Mycotoxins

Twenty gram samples from each experimental design (jute and nylon fiber bags) were analyzed for aflatoxin using the method of Kellert and Spott (9). The modified method of Nowotny et al. (10) was used to screen 10-g samples for the other mycotoxins.

RESULTS

The use of clean dry nylon fiber bags proved very effective. Fermentation was observed to last 3 or 4 days. No fungal growth was noticed on the outside of the bag or on the fermented product even on day 3 before smoke treatment.

Using jute bags, fermentation lasted 5 to 6 days, and evidence of fungal contamination was obvious between days 2 and 3 of the fermentation. But when

the jute bags received smoke treatment from day 2 to the final day of fermentation, no fungal contamination was observed. Whitish threads observed on jute bags on day 3 disappeared when smoke treatment was applied. The use of plastic bowls for fermentation was highly unsuitable because the process took longer - 2 weeks.

When ogiri was smoked for 2 weeks, it had a very appealing aroma and texture. In contrast, the end product from the plastic bowl experiment lacked the characteristic ogiri aroma. When ogiri samples from both the jute and nylon fiber bags were assayed for mycotoxins, there was no evidence of contamination.

Effect of the Types of Wraps

Samples wrapped in dry leaves of the banana plant were less susceptible to fungal attack than ogiri wrapped in leaves of Newbouldia laevis. However, regular smoke treatment reduced the incidence of fungal contamination of ogiri in both types of leaf wraps. Plastic wrapped samples had no observable fungi even up to 2 weeks of incubation but were devoid of the pleasant aroma characteristic of the smoked product.

DISCUSSION

It has been clearly demonstrated in this study that the use of clean dry nylon fiber bags instead of jute bags for the fermentation and early smoke treatment of the fermenting mash contributed significantly to the exclusion of fungi and thereby reduced the risk of mycotoxin contamination during ogiri production. Further related studies on methods of improving fermentation techniques on other

products are now being considered.

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