J. Grad. Sch. Agr. Hokkaido Univ., Vol. 70, Pt. 2 : 111~127 (2002)

Production of High-Quality Oncom, a Traditional Indonesian Fermented Food, by the Inoculation with Selected Mold Strains in the Form of Pure Culture and Solid Inoculum

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(Received Jun. 18, 2001/Accepted Aug. 10, 2001)

Abstract

High-quality oncom, traditional Indonesian fermented food, was prepared by the inoculation of pure culture of several *Neurospora, Rhizopus*, and *Mucor* strains. Although the role of *Mucor* in oncom production was not known, many *Mucor* strains could produce high-quality oncom.

Solid inoculum containing each of 4 *Rhizopus*, 4 *Mucor*, and 3 *Neurospora* strains with use of cooked rice as a carrier were prepared to produce high-quality oncom. All inoculum except one contained enough amount of living cells to produce oncom after storage for 180 days at room temperature. Bandung red oncom and bogor red oncom produced by the local producer using the solid inoculum had very good quality. They were brought to the local market and well accepted by the consumers.

Oncom is a traditional Indonesian fermented food that has been produced and consumed mainly in West Java. The raw materials of oncom are mainly the waste of agricultural products and there are many kinds of oncom by the combination of several molds and raw materials. The temperature of the solid substrate fermentation used for fermented foods in Southeast Asia is considered to be mesophilic because the incubation is usually carried out at room temperature in a very uniform tropical environment of 25° to 32°C. Indonesia is in a hot and humid zone where the people enjoy many traditional fermented foods such as tauco, kecap, tempeh, ragi tape, tape ketala, tape ketan, and oncom because of a stable high temperature of about 31°C. All these fermented foods except oncom are produced by a modernized process with the use of inoculum, but oncom is produced on the scale of a small cottage industry with no use of inoculum, so the quality of oncom is not always stable and sometimes cannot be guaranteed.

Part of the doctoral thesis of Dudi Djuhdia Sastraatmadja with the same title submitted to the Graduate School of Agriculture, Hokkaido University (2000)

Oncom is the cheapest product among these fermented foods because the raw materials for oncom are mainly the waste of agricultural products that still contain a high amount of protein suitable for consumers with low-incomes. Therefore the improvement of the quality of oncom should result not only in a supply of good and low-cost food with high protein content, but also in the efficient use of agricultural products. However, no effort has been made to improve the quality of oncom and to distribute the improved products. This paper deals with improving the quality of oncom by inoculation with selected superior mold strains in the form of pure culture and solid inoculum.

MATERIALS AND METHOD

I. Production of high-quality oncom by the inoculation of pure culture of *Neurospora*, *Rhizopus*, and *Mucor* strains

To find the microorganisms for producing a better quality oncom, many molds, including *Neurospora*, *Mucor*, and *Rhisopus* species isolated from traditional Indonesian fermented foods in West Java[1-4] and kept in the Microbial Collection Sub-Division, Division of Microbiology, Research and Development Center for Biology-LIPI, Bogor, were examined.

The raw materials for bandung red oncom consisted of peanut press cake, solid residue of tapioca flour, and soybean curd (15:5:1). For black oncom they consisted of peanut press cake and soybean curd (3:1). They were prepared under asceptic conditions that were not adopted by traditional oncom producers. The raw material was sterilized in an autoclave at 115°C for 15 min and, after being cooled, inoculated with a heavy suspension (OD_{540nm} 0.50) of 3-day-old culture of each strain and incubated at 27°C for 24 or 48 hours. All treatments including control were done in duplicate. The products were analyzed for texture visually[5] and with the use of Penetrometer, pH, water content, reducing sugars[6], and total and soluble nitrogen[7]. Organoleptic properties were evaluated by 15 panelists under the method of Larmond[8]. The products were assessed with numbers from 1 to 9, the smaller mark meaning the better quality.

a) The ability of *Neurospora* strains to produce bandung red oncom

Oncom produced by the fermentation of raw materials consisted mainly of peanut press cake by *Neurospora*, *Mucor*, and *Rhizopus* species is one of traditional Indonesian fermented food consumed especially in West Java. It is the only food for humans produced by fermentation with the *Neurospora* species. Thirteen *Neurospora* strains isolated from oncom collected from several places in West Java were examined for their ability to produce bandung red oncom.

b) The ability of *Rhizopus* strains to produce oncom with the raw material for bandung red oncom

It has been known that at least 4 *Rhizopus* species — *R.stolonifer*, *R.oligosporum*, *R.oryzae*, and *R.arrhizus* — participate in tempeh production, and the abilities to degrade carbohydrate, protein, and lipid in the raw materials are different depending on species. Some *Rhizopus* strains have also been isolated from black oncom[9]. Eight strains of *R.oryzae*, 7 of *R.oligosporum*, 3 of *R.arrhizus*, and 3 of *R. stolonifer* that had been isolated from traditional Indonesian fermented foods collected from several places in West Java were examined for their ability to produce oncom with the raw material for bandung red oncom. The incubation time was 48 hours at room temperature.

c) The ability of *Rhizopus* strains to produce black oncom

The texture of black oncom sold in the market is not yet compact, and the surface is not fully covered by myselia, which gives a grayish black color on the surface, because the incubation period is only 24 hours. Black oncom is not only cheaper than other oncom, but it contains a relatively high amount of protein and some other nutrients. Therefore black oncom is an important food for consumers with low incomes. The raw materials for black oncom were inoculated with the pure culture of each of 11 *Rhizopus* strains isolated from black oncom and incubated at room temperature for 48 hours, instead of the 24 hours adopted by local oncom producers.

d) The ability of *Mucor* strains to produce oncom with the raw material for bandung red oncom (1)

Although *Mucor* strains are nearly always isolated from traditional Indonesian fermented foods, it has not been known that *Mucor* strains have a specific role in these foods. This study was carried out to clarify the role of *Mucor* strains in the fermentation process of traditional Indonesian fermented foods to find a superior strain for oncom production. Thirty four *Mucor* strains were isolated from 8 kinds of traditional Indonesian fermented foods in West Java, and their abilities to produce oncom were examined.

Each sample of traditional Indonesian fermented foods collected at several places in West Java was put in a sterile plastic bag in aseptic condition. A small piece of each sample was put in sterile distilled water, and after shaking, one drop of each suspension was put on a medium agar in a Petri dish and incubated at room temperature for 2 days. The colony was separated and purified. The raw material for bandung red oncom was inoculated with each Mucor strain and incubated at room temperature.

e) The ability of *Mucor* strains to produce oncom with the raw material for bandung red oncom (2)

In this experiment, the abilities of each of the 3 strains of *Mucor javanicus*, *M.circinoloides*, and other *Mucor* species isolated from traditional Indonesian fermented foods to ferment the raw material for bandung red oncom were studied. The raw material for bandung red oncom was inoculated with each of 9 strains of *Mucor* strain described above and incubated at room temperature.

f) The ability of *Mucor* strains to ferment various leguminous seeds

In this experiment, the abilities of *Mucor* strains to ferment leguminous crops, which are the raw materials for traditional Indonesian fermented foods, were studied. Three *Mucor* strains were used to inoculate 7 kinds of cooked materials —peanut press cake, soybean, wing bean, giant ipil-ipil, flat nut, lima bean, and cow pea — which were the raw materials of 7 fermented foods.

II. Production of high-quality oncom using cooked rice as a carrier of selected mold strains

Superior strains proving capable of producing good oncom and kept in Microbiology Division, R & D Center for Biology — LIPI, Bogor, were used.

a) Preparation of inoculum

To 100 g of rice grain, 60 ml of distilled water was added in a large Petri dish and sterilized at 115°C for 15 min. After being cooled, they were inoculated with 9 ml of 7-day-old culure (OD_{540nm} 0.35) of each of 4 *Rhizopus*, 4 *Mucor* and 3 *Neurospora* strains that had already been known capable of producing high-quality oncom; they were incubated at 32°C for 3 days and further at 37°C for 5 days. They were then finely milled and the powdered inoculum weighed to 0.1 g or 5 g each, used later to produce oncom or to count the number of living cells, respectively, put in small plastic bags, and sealed. The inoculum was kept at room temperature for 180 days. From time to time during storage, it was counted for living cells and tested for the ability to produce oncom.

b) Production of oncom in laboratory

One hundred g of the raw material composed of peanut press cake, solid residue of tapioca flour, and soybean curd (15:5:1) was sterilized at 115°C for 15 min; after being cooled, the material was inoculated with 0.1 g of each powdered inoculum prepared above, incubated at room temperature for 24 hours, turned over, and incubated for 24 hours more to produce oncom. The deep-fried products were analyzed for the organoleptic values by 15 panelists by the method of Larmond.[8]

c) Counting of living cells

Powdered inoculum (5 g) was suspended in sterilized distilled water (45 ml). A series of dilutions was done to obtain 30 to 100 propagules growing on Rose-Bengal dextrose agar containing Terramycin as an antibacterial agent, and the living cells grown on the substrate were counted.

d) Production of oncom by oncom producers using inoculum of selected strains

Three kinds of oncom, oncom merah bandung (bandung red oncom), oncom merah bogor (bogor red oncom), and oncom hitam (black oncom), were prepared by local oncom producers with the use of inoculum prepared by the method described above. The raw material of bandung red oncom is a mixture of peanut press cake, solid residue of tapioca flour, and soybean curd, and the main microorganism is *Neurospora* species. Bogor red oncom is prepared also mainly by *Neurospora*, but the raw material is soybean curd only without any other material. Usually, black oncom is prepared with the same materials as bandung red oncom, but by fermentation with *Mucor* or *Rhizopus* species. Inoculum containing the pure culture of 3 *Neurospora*, 4 *Mucor*, and 4 *Rhizopus* species were prepared by the same method described above and used immediately by some local oncom producers in Bandung and Bogor. Oncom were produced by using 0. 1 g of inoculum for 100 g of raw material. The products were brought to the laboratory and analyzed for texture, compactness[5], and organoleptic values.[8]

RESULTS AND DISCUSSION

I. Production of high-quality oncom by the inoculation of pure culture of *Neurospora*, *Rhizopus*, and *Mucor* stains

a) The ability of *Neurospora* strains to produce bandung red oncom

After an incubation period of 24 hours, fermentation by *Neurospora* species was not enough to produce oncom, except for one strain. The products did not have a compact texture or an oncom aroma. Only one strain of *Neurospora* (M35 II/2) examined could change the raw material to oncom with a compact texture and an oncom aroma after 24 hours of incubation (Table 1). When the incubation period was extended to 48 hours, 8 strains of *Neurospora* species, including the one that produced a good product after 24 hours of incubation, could produce oncom, but 5 other strains could not. The loose texture of raw material changed to a compact one as the result of the growth of mold mycelia. There was no significant change in water content or pH. The total nitrogen content increased slightly, probably because of evaporation of nonnitrogen volatile compounds formed

Table 1. The physicochemical properties of bandung red oncom prepared by the inoculation of pure culture of *Neurospora* strains.

Strains	Incubation (hr)	Softness[a] (mm/ 102.4 g/ sec)	Water Content (%)	pH	Nitrogen Content (g/100 g dried)		Organoleptic Value[b]
					Total	Soluble	
Neurospora sitophila							
M35II/2	24	3.5	67.9	6.8	5.4	0.80	2.60
〃	48	7.8	69.3	7.1	5.4	0.70	2.70
Neurospora							
Ns.1	48	6.9	68.8	7.1	5.2	1.58	
Ns.2	48	5.6	69.8	6.6	5.3	1.00	
Ns.3	48	5.1	70.2	6.5	4.2	0.58	
Ns.4	48	6.7	67.4	7.1	4.6	1.29	
Ns.5	48	6.6	67.4	7.1	4.8	1.45	
Ns.6	48	5.1	68.5	7.1	5.1	1.40	
Ns.7	48	5.4	72.0	6.0	5.1	1.45	

The average water content, pH, contents of total nitrogen and soluble nitrogen of the raw material were 67.3%, 6.4, 4.6 (g/100 g), and 0.63 (g/100 g), respectively.

[a] : Measured by penetrometer with 102.4 g of weight load.

[b] : Measured by "Hedonic Scale Score" with values from 1 to 9, one for extremely liked products and 9 for extremely disliked ones.

during fermentation. Soluble nitrogen content increased in different degrees, depending on each species.

b) The ability of *Rhizopus* strains to produce oncom with the raw material for bandung red oncom

It was shown that 4 *Rhizopus* species examined had nearly the same abilities to produce oncom as they did to produce tempeh. All the substrates changed in different degrees, depending on each strain. The growth of mold was heavy, and

Table 2. The physicochemical properties of oncom prepared by the inoculation of pure culture of strains of four *Rhizopus* species on the raw material for bandung red oncom.

Strains	Softness[a)](mm/ 102.4g/ sec)	Water Content (%)	Com-pact-ness[b)]	pH	Soluble Nitrogen (g/100 g dried)	Redu-cing Sugar (mg/g)	Organo-leptic Value[c)]
R. oryzae							
M40I/1	2.95	58.1	++++	5.3	0.68	73.5	3.30
M39I/1	2.40	49.9	++++	5.8	0.58	65.5	3.23
M19LII/3	2.38	68.7	++++	6.6	1.04	64.0	4.23
TR32	2.05	52.5	++++	6.1	0.64	65.0	3.30
UD79/2	1.77	52.6	++++	6.2	0.56	70.2	3.00
UD68	2.40	53.2	++++	6.1	0.63	65.2	3.30
UD67/1	2.26	55.3	++++	6.2	0.66	57.5	3.66
UD58	2.70	52.8	++++	6.3	0.61	70.5	3.03
R. oligosporus							
F.21.1.1a	4.93	68.6	++++	6.4	1.35	46.5	3.77
F.16.2.2a	3.15	65.8	++++	6.4	1.25	57.5	3.57
M6L/2	2.96	67.9	++++	6.0	0.76	61.4	3.54
F.35.1.1	2.68	63.7	++++	5.6	1.00	26.0	2.90
UD65	2.45	66.6	++++	5.9	1.07	32.2	2.90
UD50	3.19	68.2	++++	6.4	1.05	42.4	3.67
UD98	2.92	56.7	++++	6.1	0.64	30.5	4.00
R. arrhizus							
UD60	2.54	64.4	++++	6.2	1.06	40.2	4.00
UD100	2.51	55.3	++++	6.4	0.80	50.2	3.47
UD77	2.51	66.6	++++	6.1	0.99	30.2	3.87
R. stolonifer							
R1 II/1	2.35	56.5	++++	6.1	0.40	10.2	4.50
R1 II/2	2.36	55.6	++++	6.0	0.45	11.4	4.45
R2 I/1	2.45	56.4	++++	6.1	0.50	12.3	5.00

Incubation time was 48 hrs.
The average contents of water and soluble nitrogen were 63.3% and 0.28 (g/100 g).
[a)] : Measured by penetrometer with 102.4 g of weight load.
[b)] : + Poor mold growth, loose texture.
++ Moderate mold growth, moderately compact and soft texture.
+++ Good mold growth, finely compact and soft texture.
++++ Excellent mold growth, excellent soft texture.
[c)] : Measured by "Hedonic Scale Score" with values from 1 to 9, one for extremely liked products and 9 for extremely disliked ones.

the texture of the products was compact and soft (Table 2). The average soluble nitrogen content of the products inoculated with *R.oligosporus* was highest, followed by *R.arrizus* and *R.oryzae*. *R.stolonifer* produced the least amount of soluble nitrogen. It has been reported that *R.oligosporus* has a very high ability to degrade protein[10]. The average reducing sugar content of the products inoculated with *R. oryzae* was highest, followed by *R.oligosporus* and *R.arrhizus*. *R.stolonifer* also produced the least amount of reducing sugars. The organoleptic values of the products by *R.oryzae*, *R.oligosporus*, and *R.arrhizus* were from 3.00 to 4.00, which were values well received by panelists[8]. The products by *R.stolonifer*, which had the least amount of soluble nitrogen and reducing sugars contributing to taste, had poorer organoleptic values compared with those produced by the other three *Rhizopus* species.

c) The ability of *Rhizopus* strains to produce black oncom

Many *Rhizopus* strains had the abilities to produce products suitable for consumer tastes. The water content decreased and the product became harder during incubation (Table 3). The extension of incubation time to 48 hours from 24 hours adopted by oncom producers gave products with compact and soft texture covered by a heavy growth of mycelia. Soluble nitrogen content increased in different degrees, depending on strain. The tastes of the products were well

Table 3. The physicochemical properties of black oncom prepared by the inoculation of pure culture of *Rhizopus* strains.

Strains	Softness[a] (mm/ 102.4 g/ sec)	Water Content (% in dried)	Com-pact-ness[b]	Soluble Nitrogen (g/100 g dried)	Organo-leptic Value[c]
S_1II/2	3.09	53.3	++++	0.74	3.03
S_1III/1	3.23	51.9	++++	0.50	3.86
R_1II/1	2.54	40.5	++++	0.37	3.00
R_2I/1	2.66	38.3	++++	0.49	3.30
R_2III/1	2.62	37.8	++++	0.45	3.33
R_3I/1	3.05	40.6	+++	0.48	3.15
R_3II/2	2.35	30.2	++++	0.53	3.37
R_4I/1	2.53	41.5	++++	0.49	3.63
R_4I/2	2.33	33.1	+++	0.46	2.73
R_5I/1	2.86	35.8	++++	0.46	3.05
S_1X	2.61	36.3	+++	0.53	3.47
Control		45.3		0.37	

Incubation time was 48 hrs.
[a] : Measured by penetrometer with 102.4 g of weight load.
[b] : + Poor mold growth, loose texture.
++ Moderate mold growth, moderately compact and soft texture.
+++ Good mold growth, finely compact and soft texture.
++++ Excellent mold growth, excellent soft texture.
[c] : Measured by "Hedonic Scale Score" with values from 1 to 9, one for extremely liked products and 9 for extremely disliked ones.

accepted by panelists with the best organoleptic score of 2.73 of the Larmond scale. The products had a specific aroma of black oncom similar to that of tempeh.

d) The ability of *Mucor* strains to produce oncom with the raw material for bandung red oncom (1)

Thirty-four *Mucor* strains were isolated from eight kinds of traditional Indonesian fermented foods collected in several places in West Java (Table 4). Not all traditional Indonesian fermented foods contained *Mucor* strain. The *Mucor* species most frequently isolated was *Mucor javanicus*, which has been shown to have relatively high amylolytic, proteolytic, and lipolytic activities and to produce oncom and tempeh of good quality[11]. Other Mucor species isolated were *M.circinolloides* (3), *M.Rouxii* (1), *M.inaequisiporus* (1), and *Mucor* species unidentified (20). Generally, Mucor strains can produce oncom and tempeh with better taste and aroma than those by *Rhizopus* strains[12]. Thirty-two of 34 *Mucor* strains could grow on the raw material for bandung red oncom and produce the products in 24 hours of incubation (Table 5). The texture of the products was soft and compact, and the surface was covered by grown mold. Although total nitrogen content decreased slightly, soluble nitrogen and reducing sugar content increased. When incubation time was extended to 48 hours, all 34 *Mucor* strains could produce oncom with better texture compared with those with 24 hours of incubation. It was proven that although *Mucor* strains are not the main mold in fermented foods, they can produce a product with the same quality as those by main molds.

e) The ability of *Mucor* strains to produce oncom with the raw material for bandung red oncom (2)

All 3 *M.javanicus* strains produced oncom with good mold growth and a finely compact and soft texture after 24 hours of incubation (Table 6). The tastes of the three products were similar and accepted by panelists with a Larmond

Table 4. Presence of *Mucor* in several traditional Indonesian fermented foods in West Java

Places	Traditional Indonesian Fermented Foods								
	Tauco	Kecap	Tempeh	Ragi tape	Red oncom	Tape ketala	Black oncom	Tape ketan	Total
	Numbers of *Mucor* strains Isolated								
Jakarta	-	-	-	-	-	-	1	-	1
Bogor	-	2	4	-	1	-	-	-	7
Cianjur	8	2	1	-	-	2	-	-	13
Sukabumi	-	1	1	-	1	-	-	-	3
Bandung	-	-	-	2	-	-	-	-	2
Garut	-	-	-	2	2	1	-	-	5
Karawang	-	2	-	1	-	-	-	-	3
Total	8	7	6	5	4	3	1	0	34

Table 5. The physicochemical properties of oncom prepared by the inoculation of pure culture of 34 strains of *Mucor* isolated from traditional Indonesian fermented foods in West Java on the raw material for the bandung red oncom.

Incubation (hr)		Softness[a] (mm/ 102.4 g/ sec)	Water Content (% in dried)	Compactness[b]	pH	Nitrogen Content (g/100 g dried)		Reducing Sugar (mg/g)
						Total	Soluble	
24	32/34[c]	4.65	65.9	++	5.7	2.0	0.26	35.6
	Control[d]		67.3		5.9	2.1	0.04	16.0
48	34/34	3.48	67.2	++++	5.8	2.1	0.15	40.2
	Control[d]		69.0		5.8	2.2	0.05	21.9

Averaged values of 32 and 34 products for 24 and 48 hr incubation, respectively.
[a] : Measured by penetrometer with 102.4 g of weight load.
[b] : + Poor mold growth, loose texture.
++ Moderate mold growth, moderately compact and soft texture.
+++ Good mold growth, finely compact and soft texture.
++++ Excellent mold growth, excellent compact and soft texture.
[c] : Strains that produced oncom/strains examined.
[d] : Uninoculated raw material.

scale of less than 3. The extension of the incubation period to 48 hours caused more change in the substrate. The products had better mold growth, more soluble nitrogen and reducing sugars, a more compact and soft texture, and better organoleptic properties, showing that the products were liked more than those with only 24 hours of incubation. Two strains of M. *circinolloides* formed oncom with good mold growth, a finely compact and soft texture, and an organoleptic value of 2.9 and 3.0 Larmond scale, respectively, but the product by one strain was slightly inferior with moderate mold growth and a moderately compact and soft texture with an organoleptic value of 3.1. All three products were accepted by the panelists. The soluble nitrogen content of the products by *M.circinolloides* was lower than those by *M.javanicus*, and one *M.circinolloides* strain gave the lowest value. After 48 hours of incubation, the mold growth and texture were improved. Reducing sugar content of the products by all three strains of *M.circinolloides* became higher than of those by *M.javanicus*. Organoleptic values of the products by 2 strains of *M.circinolloides* were improved, but the value of the product by one strain, which produced the worst product after 24 hours of incubation, was not. Three strains of other Mucor species produced good oncom with finely compact and soft texture and good mold growth after 24 hours and 48 hours of incubation. The reducing sugar and soluble nitrogen content of the products prepared by this group were lower than those by *M.javanicus* and *M.circinolloides* though the growth of mold was good. Organoleptic values were comparable to those by *M. javanicus* and *M.circinolloides*, but the value became worse by an extension of incubation time to 48 hours. The extension improved the organoleptic values of 4 *Mucor* strains, but those of one strain each of *M.javanicus* and *M.circinolloides*

Table 6. The physicochemical properties of oncom prepared by the inoculation of pure culture of strains of two *Mucor* species on the raw material for the bandung red oncom.

Strains	Incubation (hr)	Softness[a] (mm/ 102.4 g/ sec)	Compactness[b]	Soluble Nitrogen (g/100 g dried)	Reducing Sugar (mg/g)	Organoleptic Value[c]
M.javanicus						
M.3.8	24	2.75	+++	0.44	33.0	2.90
	48	2.51	++++	0.52	48.0	2.60
M.4.8	24	3.29	+++	0.61	33.0	2.90
	48	2.19	++++	0.71	48.0	2.80
M.5.0	24	3.37	+++	0.62	25.0	2.70
	48	2.40	++++	0.62	58.0	2.70
M.circinolloides						
M.2.8	24	2.46	+++	0.44	25.0	3.00
	48	2.33	++++	0.54	59.0	2.80
M.7.0	24	2.54	+++	0.38	40.5	2.90
	48	2.50	++++	0.44	59.8	2.70
M.3.2	24	4.02	++	0.16	40.5	3.10
	48	4.00	+++	0.16	65.5	3.10
Mucor species (not identified)						
M.2.1	24	4.02	+++	0.13	21.0	2.25
	48	3.96	++++	0.15	18.0	2.30
M.2.7	24	3.83	+++	0.21	9.0	3.00
	48	3.25	++++	0.21	19.5	3.10
M.2.6	24	5.51	+++	0.16	15.0	3.00
	48	4.07	++++	0.18	26.0	3.05

a) : Measured by penetrometer with 102.4 g of weight load.
b) : + Poor mold growth, loose texture.
++ Moderate mold growth, moderately compact and soft texture.
+++ Good mold growth, finely compact and soft texture.
++++ Excellent mold growth, excellent compact and soft texture.
c) : Measured by "Hedonic Scale Score" with values from 1 to 9, one for extremely liked products and 9 for extremely disliked ones.

were not changed, and those of 3 other strains became worse. This experiment revealed that *Mucor* strains could usually produce oncom with good texture and organoleptic properties with the raw material for bandung red oncom, and some of the strains could be used as superior strains to produce oncom. It has been reported that some *Mucor* strains produce oncom with higher soluble nitrogen content and better texture and taste than those by *Rhizopus* species. To develop and market the products fermented by *Mucor* strains, it is essential to obtain superior strains and to be accepted by consumers. When the panelists determined that the new products fermented by *Mucor* strains can be accepted by consumers, the next issue was how to produce the products with high and constant quality, and it is important to use the inoculum containing superior strains to achieve that. More investigations should give us information about the role of *Mucor* strains in the traditional and modern fermentation industry.

f) The ability of *Mucor* strains to ferment various leguminous seeds

One *Mucor* strain could grow on various seed materials. Two of 3 *Mucor* strains grew on all substrates and produced products with a finely compact and soft texture covered with a good growth of mold after 24 hours of incubation (Table 7). After 48 hours of incubation, all 3 *Mucor* strains produced products with an excellent compact and soft texture covered with excellent growth of the mold.

There are several kinds of oncom, that is, oncom merah bandung (bandung red oncom), oncom merah bogor (bogor red oncom), and oncom hitam (black oncom). The raw material of bandung red oncom is a mixture of peanut press cake, solid residue of tapioca flour, and soybean curd. Since the ratio of each raw material varies with producers, which are usually small-scale manufacturers working from their homes, a variety of bandung red oncom with different tastes and nutritional values is produced. The main microorganisms to produce bandung red oncom are *Neurospora* species, such as *N.intermedia*[13], *N.sitophila*[14] and *N. crassa*[15], and the cakes covered with massive coat of living conidia are sold in markets. The thicker the conidial layer, the higher the commercial value of the product. They are called red oncom because of the glistening orange color of the conidia of the microorganisms. Bogor red oncom is also mainly prepared by *Neurospora*[13] as bandung red oncom, but the raw material of bogor red oncom is soybean curd only, with no other materials. Soybean curd is fermented to produce oncom or tempeh by *Neurospora* or *Rhizopus* species, respectively. Although bandung and bogor red oncom are fermented by several *Neurospora* species, *N. intermedia* is the predominant one with characteristically bright yellow-pink conidia[13].

The main raw material of black oncom is peanut press cake. Other materials added to peanut press cake depend on the producers. Usually black oncom is

Table 7. The ability of three Mucor strains to ferment leguminous seeds.

Strains	Incubation (hr)	Leguminous Seeds						
		Peanut press cake	Soy bean	Wing bean	Giant ipil-ipil	Flat nut	Lima bean	Cow pea
I	24	+++[a]	+++	+++	+++	+++	+++	+++
	48	++++	++++	++++	++++	++++	++++	++++
II	24	+++	+++	+++	+++	+++	+++	+++
	48	++++	++++	++++	++++	++++	++++	++++
III	24	+++	+++	++	++	++	++	++
	48	++++	++++	++++	++++	++++	++++	++++

[a] : + Poor mold growth, loose texture.
++ Moderate mold growth, moderately compact and soft texture.
+++ Good mold growth, finely compact and soft texture.
++++ Excellent mold growth, excellent compact and soft texture.

prepared with the same materials as bandung red oncom, but by fermentation with *Mucor* or *Rhizopus* species. The growth of mycelia on the surface of black oncom is poor and cannot give a clean color of the conidia. Bandung red oncom and bogor red oncom are produced in the Bandung area and in the Bogor and Jakarta area, respectively; black oncom is produced only in Bogor.

Mucor species have been found in some traditional fermented foods[16,17], and they could produce oncom with qualities similar to or even higher than the products fermented by the *Neurospora* or *Rhizopus* species. Popular traditional fermented foods such as tempeh and oncom are produced on a small scale by old-fashioned methods that lack sufficient asceptic control procedures. Most solid fermentation using molds usually starts at acidic pH, which increases or decreases when the substrate is protein rich or starch rich, respectively. A fair part of carbohydrates, proteins, and lipids in the raw materials is hydrolyzed and solubilized to make the products much more digestible than the raw materials.

The solid state fermentation is usually carried out at room temperature in a very uniform tropical environment of about 25° to 32°C in Southeast Asia. Although the traditional practice with a thin layer of substrate generally less than 3.5 cm thick allows some aeration to the center of the fermenting cake, the growth of fungi at the center is restricted because fungi are strict aerobes and spore formation requires more aeration, as indicated by sporelation of conidia of *Neurospora* restricted on the surface of the oncom cake. The stacking of the trays with wide gaps during fermentation of concom ensures adequate aeration. As described above, most traditional fermented foods are still produced by traditional methods in a small cottage industry, where specific inoculation is not used, but the powdered conidia of old oncom or cut-dried old oncom is spread over the surface of the substrate. A more simple method, natural inoculation, has also been used. The substrate is left in an open space where it is inoculated by spores living in the space. This simple method has frequently been used by oncom producers because *Neurospora* is predominant in an oncom-producing room. The main problem in developing and spreading the improved method, using the selected inoculation, is how to persuade the traditional oncom producers to try using it. Traditional oncom producing is a cottage industry, and oncom producers are conservative. It is not easy for them to accept any change in the method they have employed to produce oncom. On the other hand, tempeh inoculum is easily obtained in any market and has been used by nearly all tempeh producers.

The best development in varieties of cultures and forms of inoculum has been seen in Japan, where homemakers can buy, for instance, the necessary inoculum of *Bacillus natto* to make natto, which is soybean fermented by the bacteria. Baker's yeast is also a classical household commodity regularly used in the Western world.

The production of fermented foods in a small-scale industry using inoculum produced by advanced technology is considered to be the ideal combination for traditional fermented food production. Although traditional fermentation industries operate on a small scale and producers are conservative, they can be guided in a positive direction to create a variety of inexpensive products with high nutritional value.

II. Production of high-quality oncom using cooked rice as a carrier of selected mold strains

Although the numbers of living cells in all inoculum decreased during storage for 180 days, 10 of 11 inoculum still contained more than 10^4/g of living cells after 180 days and could produce oncom with different degrees of taste (Table 8). But living cells in one inoculum (*Mucor* strain M5 II/4) decreased rapidly to 10^3/g, and the inoculum could not produce oncom. Although the amounts of propagules of more than 10^3/g inoculum were critical to form the products, the number of living cells in the inoculum did not necessarily correlate to the ability of the inoculum to produce oncom. This is because the ability of inoculum to produce oncom depended not only on the number of living cells, but also on the size of propagules, which influenced the velocity of the strain to spread and cover the surface of substrate. The inoculum of three strains — each of *Neurospora*, *Rhizopus*, and *Mucor* species — could produce oncom with very good taste (organoleptic scores of 2.71, 2.73, and 2.60, respectively) after 180 days of storage. Five inoculum produced acceptable oncom (3.00-3.71), but two produced poor products (4.20).

Oncom were produced by local oncum producers using the inoculum of pure culture.

- Bandung red oncom

One bandung red oncom produced with the use of inoculum of a *Neurospora* strain was a good one with heavy mold growth and a compact and soft texture (Table 9), which was liked by the panelists (organo-leptic score 2.80). The products were brought to the local market in Bandung to learn if the consumers who usually buy oncom produced by the local producer would accept the product. Because all the products were sold and the texture and taste were well accepted, the producer decided to use the inoculum for the next production. However, he would not use the new inoculum; he used the old oncom prepared by this inoculum containing a selected strain of *Neurospora*. But some other producers did not want to change the traditional method because they considered that their method could make good oncom and the products sold well. It has been proven that the *Neurospora* strain producing a good oncom has a better ability to degrade the raw materials of bandung red oncom than others do.[19,20]

- Bogor red oncom

The inoculum of *Neurospora* strain that could produce bandung red oncom with high quality also produced very good bogor red oncom (Table 9). The texture and taste of the product were better than those of local oncom. The local producer again tried to make oncom by using the same inoculum, and all the

Table 8. Changes in the amounts of living cells in inoculum containing selected *Neurospora*, *Rhizopus*, and *Mucor* strains during storage and the organoleptic values of oncom prepared by those inoculum.

Strains	Storage Period of Inoculum (Days)											
	0		7		14		30		90		180	
	Amount of Living Cell/g Inoculum	Organo-leptic Value[a]	Amount of Living Cell/g Inoculum	Organo-leptic Value[a]	Amount of Living Cell/g Inoculum	Organo-leptic Value[a]	Amount of Living Cell/g Inoculum	Organo-leptic Value[a]	Amount of Living Cell/g Inoculum	Organo-leptic Value[a]	Amount of Living Cell/g Inoculum	Organo-leptic Value[a]
Rhizopus												
F.21.1.1a	2.80×10^4	2.96	2.20×10^4	3.13	2.00×10^4	3.65	1.90×10^4	2.67	1.85×10^4	–	1.45×10^4	3.30
F.16.2.2a	4.50×10^4	3.40	3.50×10^4	3.06	2.60×10^4	3.86	1.90×10^4	2.67	1.60×10^4	–	1.40×10^4	2.73
TR32	9.60×10^4	4.00	9.60×10^4	3.06	9.20×10^4	3.26	8.80×10^4	3.06	8.38×10^4	–	8.12×10^4	4.20
F.23.2.2a	5.50×10^4	–[b]	4.40×10^4	–	3.50×10^4	–	3.20×10^4	–	2.00×10^4	3.86	1.30×10^4	3.60
Mucor												
F.15.2.1a	1.20×10^8	2.13	1.10×10^8	2.73	1.05×10^8	2.53	9.50×10^7	2.67	7.50×10^7	–	6.00×10^7	3.06
M34 II/3	8.70×10^6	2.66	3.50×10^6	2.75	2.33×10^6	2.53	1.95×10^6	2.75	1.30×10^6	–	1.10×10^6	4.20
M5 II/4	3.93×10^4	–	3.50×10^4	–	1.90×10^4	–	1.10×10^4	–	6.50×10^3	2.46	1.02×10^3	
F.24.1.2	1.54×10^5	–	1.45×10^5	–	1.25×10^5	–	5.30×10^4	–	3.60×10^4	3.00	2.80×10^4	2.60
Neurospora												
M46 II/1	9.90×10^4	–	8.50×10^4	–	7.90×10^4	–	5.50×10^4	2.86	4.90×10^4	3.20	3.50×10^4	3.71
M22 II/3	1.40×10^5	–	8.00×10^4	–	5.80×10^4	–	4.40×10^4	3.13	3.40×10^4	3.06	3.10×10^4	3.00
M35 II/1	7.50×10^6	–	6.40×10^6	–	5.90×10^6	–	4.50×10^6	3.07	3.80×10^6	3.06	3.20×10^6	2.71

[a] : *Organoleptic values measured by "Hedonic Scale Value" with values from 1 to 9, one for extremely liked products and 9 for extremely disliked ones.*
[b] : Not determined.

Table 9. The physicochemical properties of oncoms produced at local oncom producers by using the inoculum containing selected mold strains with the raw materials for bandung red oncom, bogor red ncom and black oncom.

Strain	Raw Material for								
	Bandung Red Oncom			Bogor Red Oncom			Black Oncom		
	Amount of Living Cell/g Inoculum	Texture[a]	Organoleptic Value[b]	Amount of Living Cell/g Inoculum	Texture	Organoleptic Value	Amount of Living Cell/g Inoculum	Texture	Organoleptic Value
Neurospora									
I	8.15×10^7	++++	2.80	6.30×10^3	++++	2.75			
II	2.75×10^4	++	3.25	2.25×10^5	+++	3.20			
III	3.12×10^5	++	3.45	3.12×10^5	+++	3.39			
Local Product[c]		++++	3.01		++++	3.10			
Rhizopus									
IV							1.97×10^4	++	3.10
V							1.46×10^4	++	3.19
VI							2.11×10^4	++	4.01
Mucor									
VII							3.13×10^7	++++	2.80
VIII							5.21×10^7	++++	2.70
Local Product								++++	3.10

a) : Texture
+ Poor mold growth, loose texture.
++ Moderate mold growth, moderately compact and soft texture.
+++ Good mold growth, finely compact and soft texture.
++++ Excellent mold growth, excellently compact and soft texture.

b) : Organoleptic values measured by "Hedonic Scale Value" with values from 1 to 9, one for extremely liked products and 9 for extremely disliked ones.

c) : Oncom produced by the traditional method without use of inoculum.

products were sold without any complaint. But complaints came from producers because they considered that extra cost would be necessary to use inoculum and that they could produce oncom by the traditional method without using inoculum. The price of bogor red oncom is relatively low compared with that of bandung red oncom, and it has been produced in a small-scale cottage industry. Therefore the inoculum of a selected superior strain, which is not expensive and is easy to obtain, must be prepared and the training of producers done without a fee.

· Black oncom

All products made with use of inoculum cotaining *Rhizopus* species showed poorer growth of mold and worse texture than those of local products, but the products inoculated with inoculum of *Mucor* species had the very good texture and taste (Table 9). The results confirmed the data obtained previously that the pure culture of *Mucor* strains had better ability than *Rhizopus* species to produce oncom.[11,20] Although bandung and bogor red oncom are produced by 48 hours incubation, black oncom is produced by 24 hours incubation. Therefore, a relatively large amount of inoculum has to be inoculated every day to produce oncom with good quality. Because of this problem, local oncom producers do not want to use the inoculum to produce black oncom. The effort to improve the quality of black oncom has to be continued.

Acknowledgments

This study was partly supported by the JSPS (Japan Society for the Promotion of Science) RONPAKU PROGRAM.

A part of this report has been accepted to be published as two rapid papers in *Annual Reports of ICBiotech.*

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