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Effect of Oxygen Concentration on Deteriorative Mechanisms of Rice during Storage

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ABSTRACT

Interseed air of rice during storage is composed of various concentrations of oxygen as well as carbon dioxide, whether rice is stored in sacks or in bulk. It is believed that the oxygen concentration of the storage atmosphere has much effect on changes of rice properties during storage. Rice was stored in air and oxygen-free gases (experiment I), and in mixtures of oxygen and carbon dioxide in several different proportions (experiment II). Rice stored in low oxygen 1) showed decreases in acidity of water-extract and a great increase in reducing sugars during storage, although amylase activity was not influenced by oxygen concentration; and 2) produced some alcohol. Judging from these facts, rice components, during storage in low oxygen, must decompose by different mechanisms from that during storage in air. Production of alcohol by the anaerobic respiratory system of rice is well explained by D. L. Taylor's theory. Evaporation of alcohol may be one cause of weight loss of rice during storage.

Among many factors governing the condition of rice during storage, oxygen concentration of the storage atmosphere is believed to have an important effect on rice quality (1). When rice is stored in bulk without aeration, the storage atmosphere becomes oxygen-deficient or oxygen-free as the result of respiration of the rice or microorganisms living on it. The same conditions may also arise when brown or milled rice is stored in cans, polyethylene bags, or even paper bags. Milner *et al.* (2,3) have reported on changes of carbon dioxide and oxygen concentrations of interseed air during storage of soybeans and wheat.

Experiments were performed to evaluate the effect of different atmospheres on changes in rice composition during storage and thus to obtain some information on comparative mechanisms of deterioration. In the first experiment, rice was stored in air and in oxygen-free gases; in the second, in mixtures of oxygen and carbon dioxide of several different proportions.

MATERIALS AND METHODS

In experiment I, brown rice of Mihonishiki variety, a short-grain rice produced in Japan, was used. Moisture content was regulated to about 16% before storage, and the rice was stored for 1 year at the natural temperature (14.7°C. on the average, about 33°C. at the highest and –2°C. at the lowest), in containers filled with 1) nitrogen, 2) carbon dioxide, and 3) air.

Properties measured before and after storage were: moisture content, acidity of water-extract, fat acidity, reducing sugar content, peroxide content, alcohol content, and number of fungi living on the rice. The data describing these properties were reported on dry basis.

Moisture was determined as the loss in weight after 5 hr. of drying at 105°C. at atmospheric pressure. Acidity of water-extract was determined by the AOAC method (4), and fat acidity by the AACC method (5).

Peroxide content was determined according to the method of Hills and Thiel (6). This consists in measurement of the yellow color produced by peroxides in a benzene extract of ground rice on addition of ferrous chloride and ammonium thiocyanate. The intensity of the color is determined by the spectrophotometer at 490 mμ.

Alcohol content was determined by the method of Reid and Truelove (7). Ground brown rice is heated in the dry state in a test tube in an oil bath at 135°C. for 25 min. The distillate is collected in water in a trap kept at 0°C. A red color is formed by adding ceric ammonium nitrate to it. The intensity of color is determined by the spectrophotometer at 530 mμ. The method will permit determination of 0.01% alcohol in the trap water with a probable error of 10%.

The numbers of fungi were counted by culturing a water suspension of ground brown rice on malt-salt-boric agar containing rose bengal as the bactericide at the rate of 70 mg. per liter (8,9). After several days, the colonies were counted.

In experiment II, brown rice of Hatsunishiki variety (a short-grain rice produced in Japan) was stored at 23°C. for 200 days in containers filled with mixtures of oxygen and carbon dioxide of various proportions: 1) pure carbon dioxide; 2) a mixture of carbon dioxide and air at 4:1; 3) a mixture of carbon dioxide and air at 1:1; 4) air; and 5) oxygen-enriched air, containing twice as much oxygen as in normal air.

Properties measured before and after storage were: moisture content, germination percentage, acidity of water-extract, alcohol content, alpha-amylase activity, beta-amylase activity, and the numbers of fungi, bacteria, and actinomyces.

Alpha-amylase activity was determined by Fuwa's blue value method (10), and beta-amylase activity by measuring reducing sugars before and after reaction. The numbers of bacteria and actinomyces were counted by using Fraenkel agar medium. Other properties were determined by the same methods as in experiment I.

RESULTS AND DISCUSSION

Results of experiment I are shown in Table I.

In comparing open-air storage with nitrogen or carbon dioxide storage, variations in moisture content and mold infection necessarily have effects in addition to the differences between aerobic and anaerobic conditions. Nevertheless, results of experiment I showed almost the same trends of changes as those of experiment II in which moisture was kept at the same level. There

TABLE I

CHANGES IN CHARACTERISTICS OF RICE DURING STORAGE IN AIR AND IN OXYGEN-FREE GASES[a]

Characteristic and Unit	Before Storage	Storage Conditions In N_2	In CO_2	In Air
Moisture content, %	16.1	16.2	16.2	14.4
Acidity of water-ext., mg. KOH/100 g.	131.5	111.7	120.7	141.1
Fat acidity, mg. KOH/100 g.	30.7	46.8	46.1	35.0
Peroxide content, μmol. O_2/100 g.	1.04	2.05	1.98	2.38
Alcohol content, mg./100 g.	5.5	46.3	55.4	5.2
Reducing sugars, mg./100 g.	258.5	369.4	388.2	281.5
Number of fungi/g.	....	33	33	9,900

[a] Results expressed on dry basis.

were differences only in degree of changes between the results of experiments I and II.

Results of experiment I show that:

Acidity of the water-extract increased in air, but decreased in nitrogen or carbon dioxide.

The increase in peroxides of rice stored in air was greater than that of rice stored in nitrogen or carbon dioxide.

Rice stored in nitrogen or carbon dioxide produced some alcohol.

The increase in reducing sugars of rice stored in nitrogen or carbon dioxide was greater than that of rice stored in air.

The number of fungi living on rice stored in air was large.

Storage results in nitrogen and in carbon dioxide were essentially the same.

Results of experiment II are shown in Table II.

The results show trends similar to those found in experiment I, but in differing degree.

Results of experiment II show that:

TABLE II

CHANGES IN CHARACTERISTICS OF RICE DURING STORAGE IN MIXTURES OF CARBON DIOXIDE AND OXYGEN IN VARIOUS PROPORTIONS[a]

Characteristic and Unit	Before Storage	Storage Conditions In CO_2	CO_2:O_2 4:1	CO_2:O_2 1:1	In Air	Oxygen-Enriched Air
Moisture content, %	15.0	15.3	15.1	15.1	15.3	15.4
Germination, %	100	0	3	9	12	7
Acidity of water-ext., mg. KOH/100 g.	157.2	142.6	137.1	139.4	234.2	199.0
Alcohol content, mg./100 g.	5.7	41.9	39.6	82.5	11.8	0
Reducing sugars, mg./100 g.	274.1	433.9	431.6	438.1	414.9	398.3
Amylase activity						
Alpha-, % decrease of light abs.	12.4	5.0	4.8	5.6	7.4	3.2
Beta-, mg. glucose produced	88	60	64	70	66	76

[a] Results expressed on dry basis.

1. Rice stored in normal air showed the highest germination percentage, and rices stored at either higher or lower oxygen levels showed lower percentages.

2. Acidity of the water-extract of rice increased in air and decreased in low oxygen during storage.

3. There was no definite difference in changes of amylase activity.

4. The increase in reducing sugars of rices stored in low oxygen was greater than that of rice stored in air.

These increases in reducing sugars and the change in amylase activity suggest a different mechanism in the starch decomposition of rices stored in air and in low oxygen.

Under aerobic conditions, when storage conditions are not good, respiration of rice is accelerated, organic acids increase, and reducing sugars decrease. In the present reducing-sugar balance of supply and consumption, reducing sugars are accumulated in rice grains despite the increase in their consumption. This suggests that the velocity of the starch decomposition to reducing sugars continues to be greater than that of the decomposition of reducing sugars.

Starch → reducing sugars → organic acids → carbon dioxide and water
Supply → | → Consumption

Under anaerobic conditions, the decomposition mechanism of starch is somewhat different. The increase in reducing sugars is larger than that of rice stored under aerobic conditions. This larger accumulation of reducing sugars is more likely caused by the decrease of reducing-sugar consumption than by the increase in starch decomposition. Under anaerobic conditions, respiration is strongly depressed and production of organic acids from reducing sugars is likewise controlled, but depression of respiration does not control the starch decomposition by amylase activities.

5. Rice stored under anaerobic conditions produces some alcohol. A part of it may be produced by enzymes of microorganisms, but most must be produced by enzymes of rice grains. D. L. Taylor, a botanist, in 1942 discovered an anaerobic respiratory system in rice alone among many varieties of cereals (11), and he proposed that rice can germinate under low oxygen concentration in water under this system. Production of alcohol during storage in low oxygen concentrations is well explained by Taylor's theory. Alcohol may largely evaporate when rice is moved to open air, which suggests one factor of weight loss of rice during storage.

Weight losses of rice during storage may be considered in three categories: 1) evaporation of water originally present as moisture in the rice grains; 2) loss of gases such as carbon dioxide caused by aerobic or anaerobic respiration; and 3) evaporation of volatile substances such as water, alcohol, organic acids, and the like which are newly produced from other substances by respiration of the rice grain and associated microorganisms.

6. Results of microbiological studies were irregular and are not shown in this paper. However, judging from these results, oxygen concentration may also have an effect on the varieties and metabolism of microorganisms living

on rice. Moreover, these differences may be effective in producing different compositional changes. A number of micrococci were found on rice stored in low oxygen; actinomyces were found only on rice stored in the mixture of equal amounts of oxygen and carbon dioxide; and a number of aspergilli and penicillia were found on rice stored in normal air.

In experiment II, there was no definite difference between rices stored in carbon dioxide, a mixture of carbon dioxide and air at 4:1, and a mixture of carbon dioxide and air at 1:1. Further experiments are required with atmospheres containing less than 50% carbon dioxide.

A question still remains whether differences are caused by oxygen concentration or carbon dioxide concentration. Although it is impossible from the present data to conclude definitely which causes more change, the concentration of oxygen is considered to have a greater effect than that of carbon dioxide. Accumulation of some carbon dioxide in interseed air is not unusual during practical storage, but no effect on compositional change has been reported.

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Literature Cited

1. ZELENY, L. Chemical, physical, and nutritive changes during storage. *In* Storage of cereal grains and their products, ed. by J. A. Anderson and A. W. Alcock; chap. II. American Association of Cereal Chemists: St. Paul, Minn. (1954).
2. MILNER, M., and GEDDES, W. F. Grain storage studies. II. The effect of aeration, temperature, and time on the respiration of soybeans containing excessive moisture. Cereal Chem. 22: 484–501 (1945).
3. MILNER, M., CHRISTENSEN, C. M., and GEDDES, W. F. Grain storage studies. VI. Wheat respiration in relation to moisture content, mold growth, chemical deterioration, and heating. Cereal Chem. 24: 182–199 (1947).
4. ASSOCIATION OF OFFICIAL AGRICULTURAL CHEMISTS. Official methods of analysis (8th ed.), Sec. 22.44. The Association: Washington, D. C. (1965).
5. AMERICAN ASSOCIATION OF CEREAL CHEMISTS. Cereal laboratory methods (6th ed.), 2.2b. The Association: St. Paul, Minn. (1957).
6. HILLS, G. L., and THIEL, C. C. The ferric thiocyanate method of estimating peroxide in the fat of butter, milk, and dried milk. J. Dairy Res. 14: 340–353 (1946).
7. REID, V. W., and TRUELOVE, R. K. Colorimetric determination of alcohols. Analyst 77: 325–328 (1952).
8. BOTTOMLEY, R. A., CHRISTENSEN, C. M., and GEDDES, W. F. Grain storage studies. X. The influence of aeration, time, and moisture content on fat acidity, nonreducing sugars, and mold flora of stored yellow corn. Cereal Chem. 29: 53–64 (1952).
9. YOSHII, H. Studies on media for the selective culturing of fungi, bacteria and yeasts living on foods. J. Food Sci. Technol. (Japan) 8: 105–113 (1961).
10. FUWA, H. A new method for microdetermination of amylase activity by the use of amylose as the substrate. J. Biochem. (Japan) 41: 583–603 (1954).
11. TAYLOR, D. L. Influence of oxygen tension on respiration, fermentation, and growth in wheat and rice. Am. J. Bot. 29: 721–738 (1942).

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