

Biogenesis of carbon dioxide for use in modified atmosphere storage of sorghum grains

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Abstract

One of the safest ways of storing cereal grains is in modified atmospheres. However, if modified atmospheres are to be used in small storage bins on farms in developing countries with minimal cost, a means is required to produce carbon dioxide and to avoid the high cost of bottled gas. To achieve this, biogenerators have been developed in which CO₂ is produced through the fermentation of waste vegetable products. This paper describes the use of saw dust, wheat bran and coffee husk as possible substrates. Saw dust allowed the production of no more than 2% CO₂ in the atmosphere while those containing wheat bran accumulated up to 19% CO₂ and those containing coffee husk accumulated up to 26% CO₂ with O₂ decreased to only 0.6%. Most CO₂ was produced by materials containing 40% water. Biogenerators containing coffee husk were connected to plastic bins containing sorghum grain with insect cages placed near the top, middle and bottom of the grain. A maximum of 21% CO₂ was recorded in the bin on the ninth day of storage, with O₂ decreased to 2%. When the biogenerators were disconnected after 14 days storage, all insects had died in the test bins but none in the control bins. Biogenerators for CO₂ production could easily be adapted for use with grain stored either in grain bins or in underground pits in rural communities in developing countries.

Introduction

Loss of food grains in developing countries after harvest is a major problem because it leads to food shortages and malnutrition. Insects are perhaps the most important agents of spoilage in stored food grains, closely followed by fungi. Both cause losses of dry matter and quality. Chemical preservation of grain is still widely practised but their toxicity and implication in ozone depletion has led to increased emphasis on non-chemical methods of preservation. Of the different methods of grain preservation in use, modified atmosphere (MA) storage has been shown to be promising in creating lethal conditions both for insects and fungi in stored grain. This method is already used to control insect infestation in large scale grain storage in Australia (Annis 1987; Ripp et al. 1990) but conditions required for the inhibition of fungi are more extreme.

In developing countries, the creation of MAs using commercial gases can be expensive, but a cheaper alternative can

be to produce them through the fermentation of plant waste materials using only the natural microflora (Paster et al. 1990). CO₂ production by peanut shells and wheat bran was compared during fermentation at different water contents in specially designed structures. Wheat bran at 40% water content produced 25% CO₂ in the atmosphere after 48 hours incubation and maintained a concentration of about 20% up to the 12th day of incubation. Peanut shells had produced slightly less CO₂ than wheat bran after 48 hours but the concentration then decreased gradually to about 12%. CO₂ production by orange peel with 80% water content, fermenting in biogenerators connected to bins containing maize grain, was similar to that by wheat bran with 35% water content after 2–7 days incubation but was significantly greater after 10 days. The CO₂ concentration in the grain was increased from less than 0.1% to about 18%, but that of O₂ was decreased from 21% to only about 10%. In experiments in Costa Rica (Paster et al. 1991), concentrations of 18.8% CO₂ and 4.7% O₂ were attained in maize grain from biogenerators containing wheat bran with 35% water content. These concentrations were sufficient to kill all insects within 9 days. Paster et al. (1991) therefore concluded that biogenerators utilising waste plant material, connected to storage bins, could be used to restrict insect damage in the small grain bins of subsistence farmers.

The present investigation aimed to test the ability of some waste plant materials commonly available in India, to produce MAs in grain bins sufficient to control storage insects.

Materials and Methods

Grain bins

Plastic bins, each holding about 25 kg of sorghum grain, were fitted with plastic tubes (0.5 cm diameter) for gas sampling at three different heights, near the top, middle and bottom of the grain bulk. Three insect cages, each containing 25 adult *Tribolium castaneum* (Herbst) 7–10 days old, were introduced at different depths in the grain bins to determine their mortality in treated and untreated bins during storage. The water content of the grain samples was determined both before and after the experiment as described by ISTA (1985).

Biogenerator construction

Biogenerators for the production of high CO₂ and low O₂ atmospheres were constructed following the design of Paster et al. (1990). A plastic container about 24 cm high and 16 cm internal diameter, with a volume of about 5 L, was fitted with a false floor of wire mesh, 4 cm above the bottom, to allow excess water to drain through to a drainage tube fixed at the bottom of the container. Another tube, also connected to the bottom of the biogenerator, transferred the gas mixture produced to a grain bin. A hole in the top, 0.5 cm diameter, allowed air access to the biogenerator to prevent anoxia.

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Substrates for biogenesis of carbon dioxide

Biogenerators were filled with sawdust (SD), wheat bran (WB), coffee husk (CH), or a 1:3 mixture of WB and SD as substrates for the production of CO₂. Lots of 800 g of each substrate, wetted to 30, 35 or 40% water content, were allowed to equilibrate at 4°C for 8 days with frequent mixing. After preliminary tests, biogenerators filled with CH or WB:SD (1:3), each containing 40% water, were selected for grain storage tests. Biogenerators containing these substrates were connected using plastic tubing (0.5 cm inner diameter) to grain bins containing about 25 kg sorghum. Each biogenerator was connected to a separate grain bin. A grain bin connected to an empty biogenerator served as a control.

Measurement of CO₂ and O₂ concentrations

Air samples were withdrawn from biogenerators and grain bins through the air sampling tubes. Instruments for measuring CO₂ (Riken, Japan) and O₂ (Gowmac, Ireland) were connected in series and air was drawn through both samplers using the peristaltic pump of the CO₂ analyser. Two replicate bins were prepared with each fermentation substrate and the experiment was repeated twice.

Results

Concentrations of CO₂ and O₂ produced in biogenerators containing four different substrates are given in Figures 1 and 2. After 2 days incubation, the CO₂ concentration in biogenerators containing wheat bran (WB) with 30% water content was significantly less than that found with 35 or 40% water content. By contrast, O₂ concentrations after 2 and 4 days incubation, with 35 and 40% water contents, were significantly smaller than those with 30% water content. Subsequently, there were no differences in CO₂ and O₂ concentrations with water content between biogenerators

containing wheat bran. The largest concentrations of CO₂ in a WB biogenerator were found after 2 days incubation with water contents of 35 and 40% and after 4 days with 35% water content.

Biogenerators containing sawdust (SD) produced maxima of 3.0, 5.0 and 5.5% CO₂ with water contents of 30, 35 and 40%, respectively, after 8 days incubation. There were no significant differences in CO₂ production with water content. The smallest O₂ concentration similarly occurred after 8 days and, again, there were no significant differences between different water contents.

Biogenerators containing coffee husk (CH) with 30, 35 and 40% water contents produced, respectively, 23, 25 and 26 % CO₂ after 2 days incubation. CO₂ production did not differ significantly with water content during incubation. After 12 days incubation, the CO₂ concentration in CH biogenerators at the three water contents ranged between 20 and 22%. O₂ concentrations decreased as CO₂ increased and, as with CO₂, there were no significant differences in O₂ concentration with water content. O₂ concentrations were never more than 2.5% during the 12 days incubation.

Largest CO₂ concentrations were found with biogenerators containing WB+SD (1:3) which reached 21% after 4 days incubation. At the same time, O₂ concentration with 40% water content declined to 1.5% after 2 days incubation. The CO₂ concentration produced by WB+SD with 40% water content did not differ significantly from that obtained with WB+SD with 30% water content after 2 and 4 days incubation, while O₂ concentrations with WB+SD at 35 and 40% water contents were significantly smaller than those with WB+SD at 30% water content.

Concentrations of CO₂ and O₂ in biogenerators containing CH and WB+SD (1:3) and in grain bins containing sorghum connected to these biogenerators are shown in Figure 3. CO₂ production in CH biogenerators were significantly greater than in those containing WB+SD except after 6 and 10 days incubation. The O₂ concentrations differed significantly

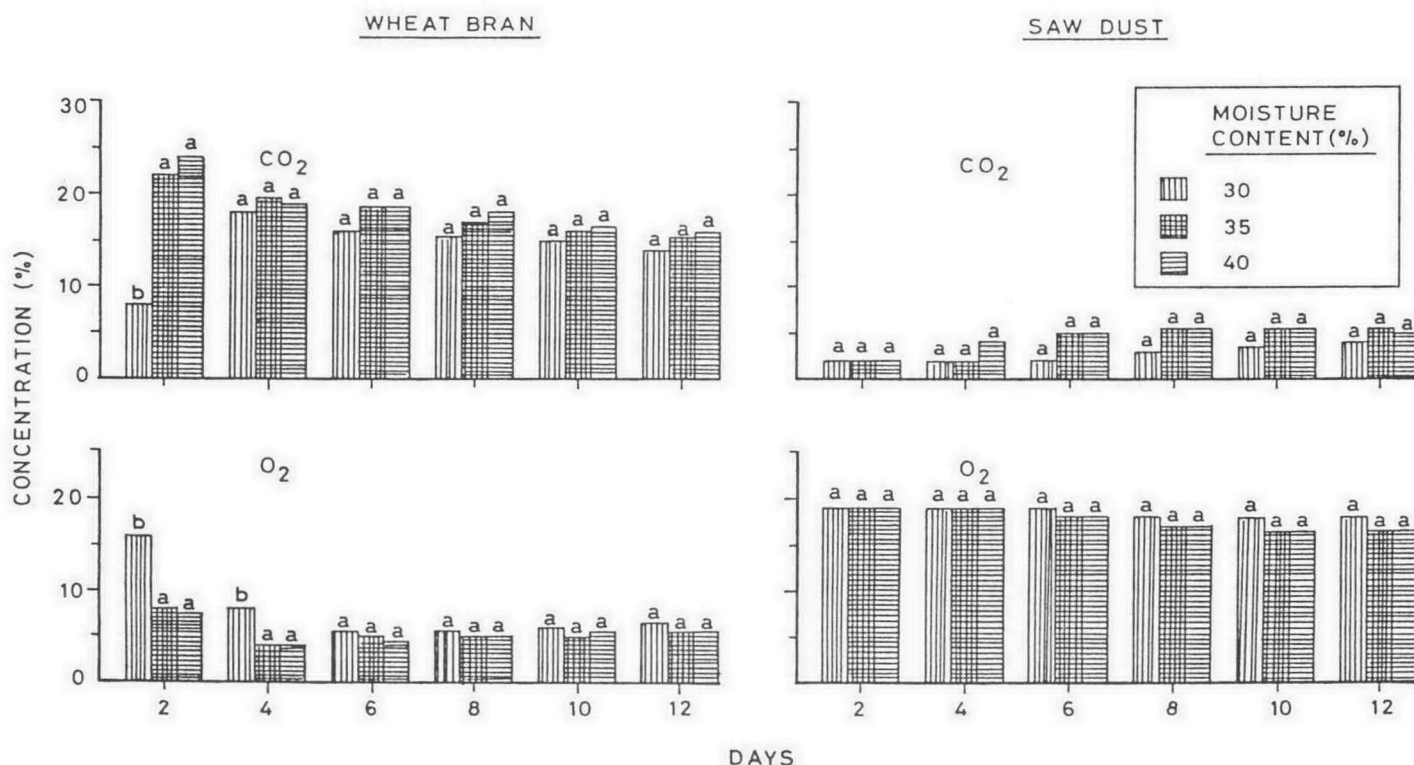


Fig. 1. Carbon dioxide and oxygen concentrations over wheat bran and sawdust at different water contents in biogenerators (within the same group, values with common letter do not differ significantly at P ≤ 0.05).

between the two substrates after 2, 10 and 12 days incubation. Concentrations of CO₂ and O₂ in the grain were smaller than those in the biogenerators to which they were connected. Concentrations of CO₂ and O₂ in grains bins connected to CH biogenerators were significantly greater than those in grain

bins connected to WB +SD biogenerators, both at the beginning and end of experiments. Control bins contained <0.1% CO₂ and 20.8% O₂.

Transfer of MAs from the biogenerators caused no change in grain water contents and there were no off-odours. All the

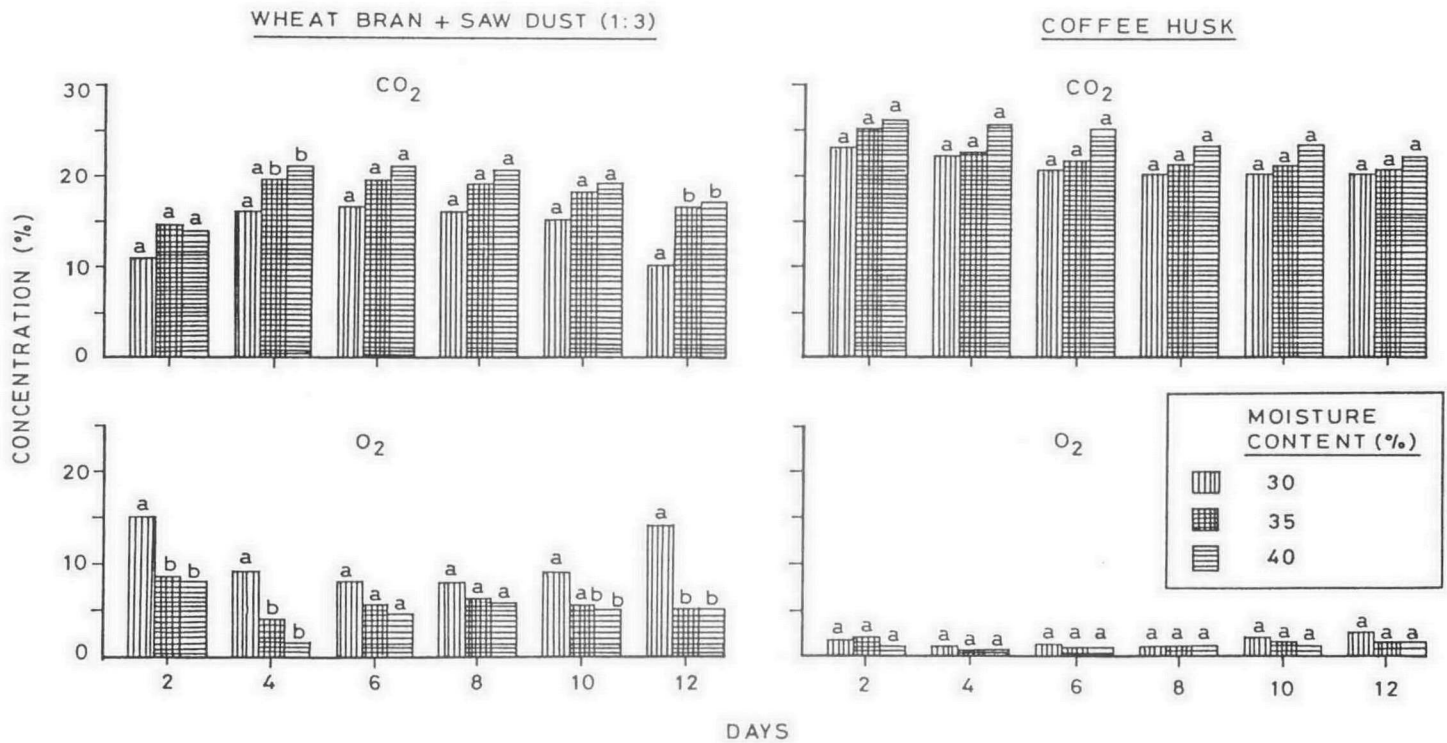


Fig. 2. Carbon dioxide and oxygen concentrations over wheat bran: sawdust (1:3) mixtures and over coffee husk at different water contents in biogenerators (within the same group, values with common letter do not differ significantly at P ≤ 0.05).

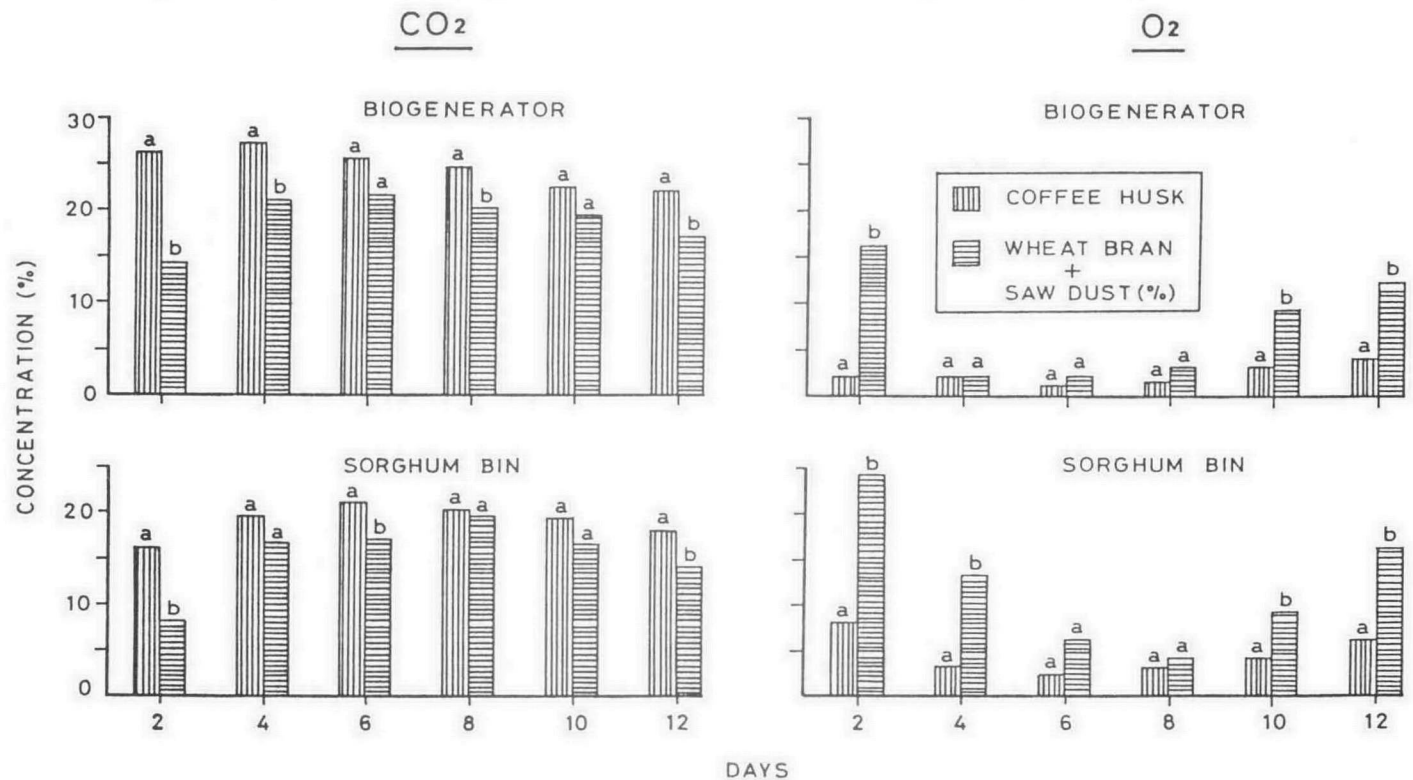


Fig. 3. Carbon dioxide and oxygen concentrations in biogenerators over wheat bran: sawdust (1:3) mixtures and in connected sorghum grain bins (within the same group, values with the common letter do not differ significantly at P ≤ 0.05).

test insects (*Tribolium castaneum*) introduced into the grain bins were killed at all three positions in grain bins connected to biogenerators but mortality in the control bins was always <2%.

Discussion

This study has confirmed that MAs can conveniently be produced by fermenting waste plant materials and that the MA produced in the biogenerators could successfully be transferred to grain bins containing sorghum. Of the three substrates used in the present study to generate MAs, CH, WB and a mixture of WB+SD produced an MA sufficient to completely kill *Tribolium castaneum* adults. Calderon and Navarro (1979) calculated that the exposure time and gas concentrations necessary to kill 95% of *T. castaneum* adults were 5 days at 22 % CO₂ and 5% O₂. The CO₂ and O₂ concentrations obtained in the present study are close to these and, with the longer exposure period (12 days), caused 100% mortality. The results also agree well with those of Paster et al. (1990–1991) and clearly indicate the potential of this system for producing MAs for small-scale storage on farms in developing countries.

Further work is needed to evaluate the effectiveness of biogenerators for controlling moulds and other insect species. However, fungi are likely to be inhibited only when there is more than 50% CO₂ in the atmosphere together with only 0.2% O₂ (Lacey 1994). Conversely, artificial inoculation of the substrate in biogenerators with fast growing microbial species could perhaps give larger concentrations of CO₂ and more rapid elimination of O₂ than the natural inoculum used in these tests. Least CO₂ was produced in the sawdust charged biogenerator and this was insufficient to be lethal to any storage insects. Significantly greater concentrations of CO₂ were produced when wheat bran and sawdust were mixed in the ratio of 1:3 than by the individual components. This could

have resulted from more vigorous microbial growth in the wheat bran and the production of enzymes which better degraded both substrates. With suitable development, this model plant-waste-material biogenerator and storage bin could be effectively used with farm-level storage structures in rural, semi-arid regions of India, even where sorghum grains are stored in the underground pits. Biogenerated MAs could easily be transferred into the pits and could significantly decrease storage losses.

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