CASSAVA AS ANIMAL FEED

Proceedings of a workshop held at the University of Guelph 18-20 April 1977

Sponsored by the International Development Research Centre and the University of Guelph

el and Michael Graham

IDRC-095e

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University of Guelph

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Cassava as animal feed: proceedings of a workshop held at the University of Guelph, 18–20 April 1977. Ottawa, IDRC, 1977. 147p. tables.

/IDRC pub CRDI/. Compilation of workshop papers on the use of /cassava/-based /feed/s — discusses methionine /amino acid/ supplementation of cassava diets; metabolic pathways and their significance in /animal nutrition/; single-cell /protein/ production (use of /bacteria/ to convert cassava into microbial protein); /animal feeding/ systems for /swine/, /poultry/ and /cattle/; /feed production/ techniques such as pelleting. /List of participants/, /statistical data/.

UDC: 636.085 ISBN: 0-88936-142-8

Microfiche Edition $1

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Fermentor Performance in Microbial Protein Production from Cassava[1]

A. G. Meiering and F. A. Azi[2]

A self-aspirating fermentor was developed on the basis of the Waldhof principle. Aeration capacity and microbial growth kinetics were analyzed for the batch and continuous flow process. Design data for a multiple stage continuous flow system were derived using the kinetic models and measured batch fermentation results in computer simulations.

Interdisciplinary research on microbial protein production from whey and cassava, a high yielding tropical root crop with very low protein but high starch content, was begun in 1971 at the University of Guelph. The whey project was aimed at the conversion of residual waste nutrients to yeast protein. The cassava project was performed under contract with the International Development Research Centre in close cooperation with the Centro Internacional de Agricultura Tropical (CIAT), Cali, Colombia. The engineering objective in the cassava project was to develop a fermentor system and a biomass harvesting system suitable for eventual use on farm or cooperative levels. This required direct cooperation with the participating research group in microbiology, which developed the microbes for the fermentation process and thus provided elementary design data for both, fermentor and separator.

Development of Fermentor

Identical fermentors, employing the Waldhof principle for the injection of air into the fermenting substrate, were used in both projects (Solomons 1969). Fig. 1 shows that the air was conveyed through the hollow shaft and impeller and then dispersed into the liquid through the action of the impeller blades, which act as a sparging vacuum pump for air (Azi et al. 1975). At the same time, it served as a liquid pump, inducing a downward flow of the substrate inside the draft tube and an upward flow between the draft tube and the vessel. The draft tube was jacketed to serve as a heat exchanger for initial steam heating of the medium as well as thermostatically controlled water cooling during fermentation. Fermentors with working capacities of 20, 120, 200, and 3000 litres were built having the same relative dimensions $D_F/H_O = 0.58$ for the fermentor vessel.

The aerating capacity of the impeller is illustrated in Fig. 2 with performance data measured at the 20 litre model. Air flow Q and power uptake P can be readily defined by the following exponential functions of rotor speed N and impeller diameter D_I (Azi 1976).

$$[1] \qquad Q = C_1 D_I^4 N^3$$

$$[2] \qquad P = C_2 D_I^4 N^{2.5}$$

The total air flow not only increased with impeller speed but also with the substrate height in the fermentor up to a point close to the equilibrium between the static potential of this height and the total potential developed by the impeller (Azi et al. 1975; Azi 1976).

The power input per litre substrate at given air flow rates increased and the rotational speed of the impeller decreased with increasing impeller diameters. A diameter of $D_I/D_F = 0.36$ provided proper substrate pumping and mixing in the vessel as well as adequate air flow rates at typical speeds between 800 and 1600 rpm and substrate filling levels between 0.66 and 0.75 H_O. Higher rotational speeds of smaller impellers improve the convective transfer of oxygen into the substrate through higher Nu-numbers and larger specific transfer areas due to smaller bubble sizes (Azi et al. 1975). At the same time, however, the shearing action of the blades is intensified, impairing microbial growth. The larger yeast cells and fungal mycelium were found to be especially sensitive to this action in the early fermentation stages.

The medium-size impeller with $D_I/D_F = 0.36$ required up to 5% more energy than the

[1]This project was financed by the International Development Research Centre in a contract with the University of Guelph. Further financial support was received in the form of an Operating Grant from the National Research Council of Canada.

[2]School of Engineering, University of Guelph, Guelph, Canada, and Engineering Division, Ministry of Agriculture and Natural Resources, Enugu, Nigeria, respectively.

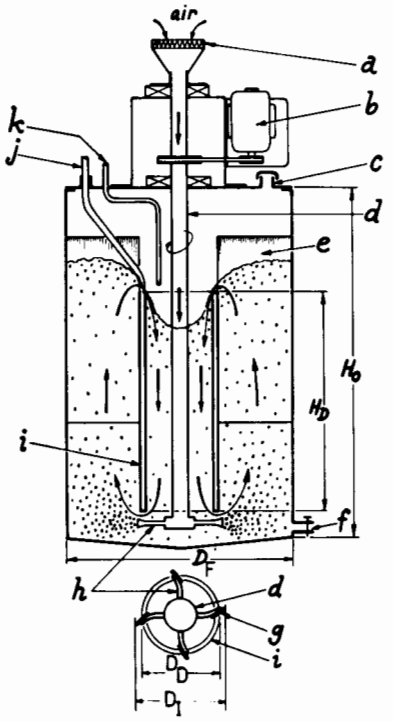

Fig. 1. Cross section through Waldhof-type fermentor: a filter; b motor and belt drive; c valve; d hollow shaft; e baffle; f outlet; g impeller; h impeller arm; i jacketed draft tube; j heating and cooling connections; and k substrate feed.

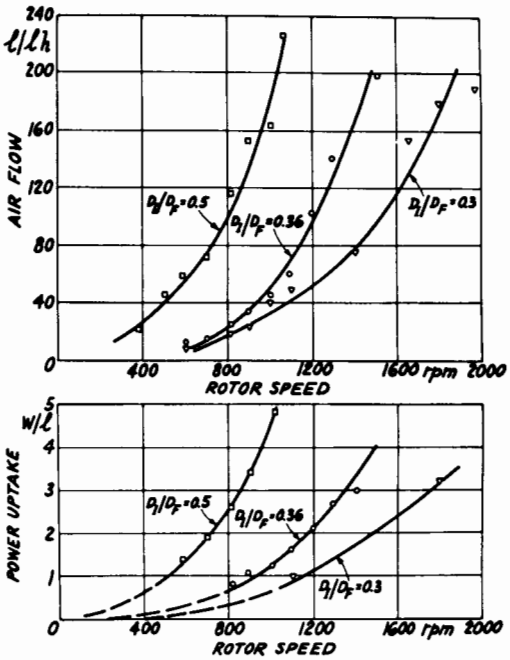

Fig. 2. Effect of impeller diameter on aerating intensity and power uptake of 20 litre fermentor. Substrate: water; fermentor dimensions: H_O = 45 cm, D_F = 30 cm, H_D = 30 cm, D_D = 10 cm (Azi 1976).

smaller one with $D_I/D_F = 0.3$, but 10–18% less than the larger one with $D_I/D_F = 0.5$. Most of the measurements needed for the selection of a proper impeller diameter were systematically taken in water and later repeated at random in various fermentation substrates (Azi et al. 1975; Azi 1976). Reasonably good agreement between these varying materials was observed. Taking all the results into consideration, an impeller diameter of $D_I/D_F = 0.36$ was chosen for the fermentation program.

Experimental Program

An asporogenous mutant (I-21A) of *Aspergillus fumigatus* was developed as a prototype for fungal cassava fermentations (Reade and Gregory 1975; Gregory et al. 1976, 1977; Gregory 1977). Between 7 and 10% inoculum was added to the prepared substrate. The cassava substrates were prepared by adding 13.3 kg of ground cassava roots to 50 litres of water. This mixture was then heated to 75 °C for 10 min by steam injection to the draft tube, to allow starch gelatinization and the elimination of some fungistatic effects (Reade and Gregory 1975). Another 50 litres of water were then added to adjust the initial carbohydrate concentration to approximately 40 g/litre and the initial substrate temperature to 45 °C. The cassava was replaced by sucrose in several experiments with fungi to minimize expenses and allow fungal biomass weights to be assayed free of cassava fibre. Concentrations of 40 and 50 g/litre were used to match whey and cassava concentrations. Minerals, phosphorus and nitrogen were added in the form of inorganic salts, urea, and corn steep liquor (see Gregory 1977).

The acidity of the cassava substrate was kept at a constant pH of 3.5 through sulfuric acid addition and the substrate temperature was kept at a constant 45 °C. These extreme growing conditions prevent the growth of contaminants and eliminate the need for an air filter. Foaming rarely occurred in initial fermentation stages and could be easily controlled through the addition of a food accept-

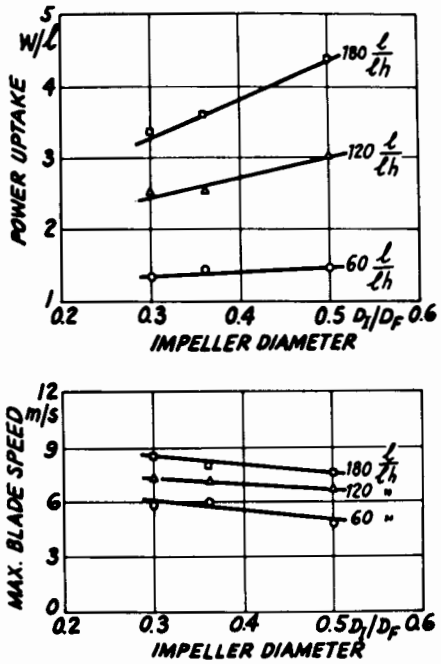

Fig. 3. Effect of impeller diameter on power requirements and impeller velocity. Substrate: water; fermentor dimensions: H_O = 45 cm, D_F = 30 cm, H_D = 30 cm, D_D = 10 cm.

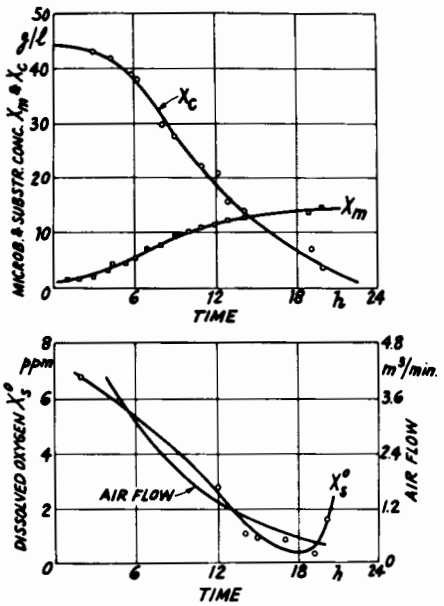

Fig. 4. Batch fermentation results of A. fumigatus I-21A in sucrose solution. Initial carbohydrate concentration 42 g sucrose/litre; substrate temperature 45 °C; impeller speed N = 800 rpm or v =13.3 m/s; fermentor size 3000 litres (Azi 1976).

able antifoam agent to whey and of small amounts of cotton seed or corn oil to the cassava substrates. Most experiments were performed with the 20 and 120 litre fermentors. They were supplemented by several runs in the 200 and 3000 litre commercial models designed for larger scale swine feed production.

Substrate samples were taken at regular intervals during all fermentation experiments and analyzed for viscosity, microbial concentration, substrate concentration, and protein yields (Reade and Gregory 1975; Gregory et al. 1976, 1977). The dissolved oxygen concentration was measured at the same time. Energy consumption of the impeller and air flow were measured in the 20 litre fermentor, which was equipped with a torque meter and a flow meter. The feed value of the biomass was determined in rat experiments (Khor 1974; Khor et al. 1976).

Fermentor Performance

The fermentor performance is characterized by the conversion rate of substrate to microbial protein (Humphrey 1974; Kihlberg 1972; Manouselis 1976; Nordstrõm 1974; Perzow 1974; Reade and Gregory 1975; Rogers et al. 1972; Weinshank and Carver 1967). The biochemical kinetics of this conversion depend on several process variables including temperature, nutrient concentration and rotor speed as the most important ones (Aiba et al. 1973; Meiering et al. 1977; Rhodes and Fletcher 1966; Tsao 1968; Weinshank and Carver 1967). An extensive theoretical analysis of the microbial reaction kinetics was performed and computer simulation programs were established for the prediction of batch and continuous flow fermentations of cassava with *A. fumigatus* I-21A (Azi 1976; Meiering et al. 1977; Monod 1942).

Measured data points and simulated reaction curves of the exponential growth phase of a typical batch fermentation experiment are shown in Fig. 4. The initial sucrose content of 40 g/litre, which compared to a similar carbohydrate concentration in the cassava slurries, was consumed after 20 h of fermentation, when a final microbial concentration of 14 g/litre was reached. The air flow decreased significantly with microbial growth, as illustrated in Figs. 4 and 5. Also, the oxygen concentration in the substrate declined sharply and

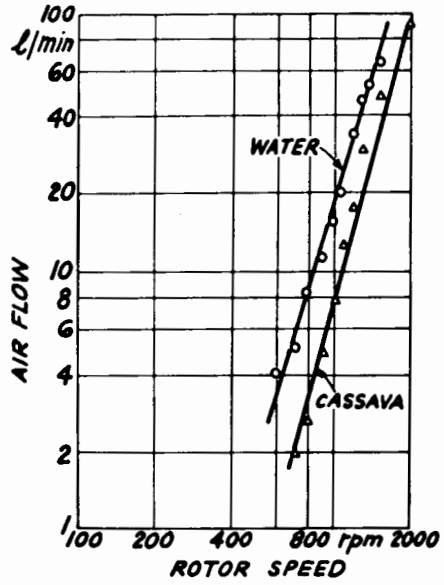

Fig. 5. Air flow through water and cassava slurry taken from 3000 litre fermentor after completed fermentation and measured on 20 litre fermentor. Fermentor dimensions: $H_0 = 45$ cm; $D = 30$ cm; $H_D = 30$ cm; $D_I/D_F = 0.36$ (Azi 1976).

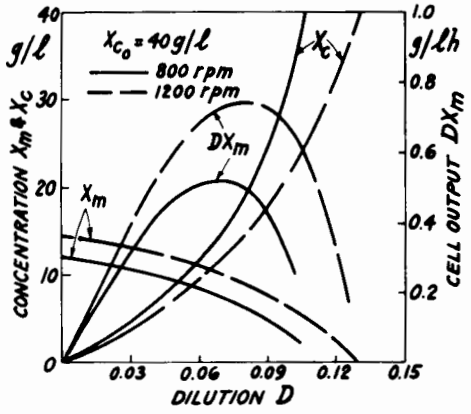

Fig. 6. Continuous flow fermentation results in single stage fermentor. Initial substance concentration 40 g sucrose/litre; substrate temperature 45 °C; impeller speed 1200 rpm or v = 20 m/s.

nearly reached depletion. The total biomass yield was 42 kg in 20 h, amounting to 0.7 kg/m^3 h^{-1} and an energy requirement of 4.97 kwh per kg biomass.

The fermentation results shown in Fig. 4 can be improved in a continuous flow system.

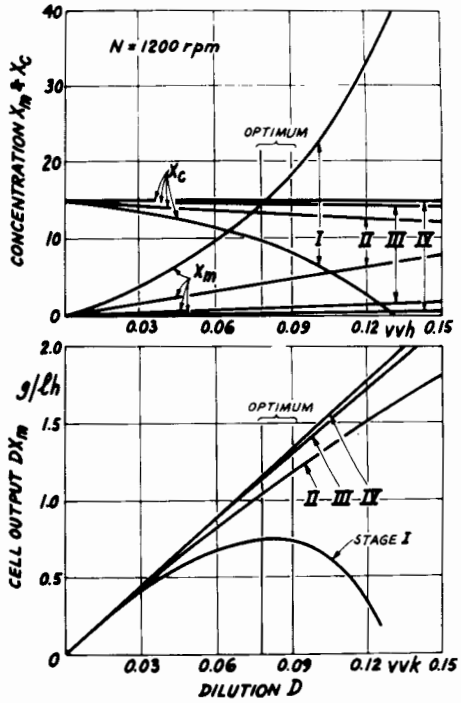

Fig. 7. Fermentation results of A. fumigatus I-21A in a multistage continuous flow system. Initial substrate concentration 40 g sucrose/litre; substrate temperature 45 °C; impeller speed 1200 rpm or v = 20 m/s (Meiering et al. 1977).

It eliminates the lag phase of microbial growth with its initial sensitivity to the shearing action of the impeller (Azi 1976). Slightly higher rotor speeds can, therefore, be used to improve the oxygen transfer and overall growth rate. Fig. 6 shows the results of continuous flow operation for the fermentor used in the batch fermentation example illustrated in Fig. 4. No improvements are achieved, if the single fermentor is operated in the same mode as in the batch process. In fact, the fermentation is incomplete at the highest cell output rate of $DX_m = 0.52$ g/litre h^{-1}, leaving a substrate concentration of approximately 14 g/litre in the effluent. The maximal cell output improves to 7.4 g/litre h^{-1} with a higher rotor speed of 1200 rpm, but approximately the same amount of substrate is lost. A dilution rate of $D = 0.09$ is required for maximal production.

Fig. 7 shows that a sequence of three fermentors is required to limit substrate losses to 1 g/litre at the maximum cell output rate of

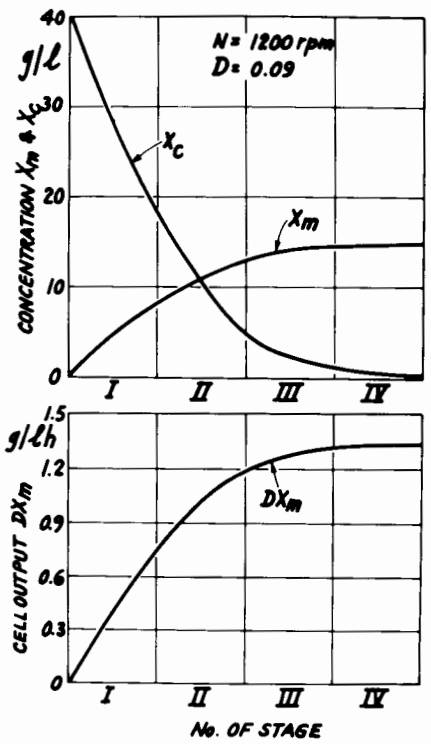

Fig. 8. Fermentation results of A. fumigatus I-21A in a four-stage continuous flow system. Initial substrate concentration 40 g sucrose/litre; substrate temperature 45 °C; impeller speed N = 1200 rpm or v = 20 m/s; dilution rate D = 0.09 in all stages (Meiering et al. 1977).

DX_m = 1.31 g/litre h^{-1} corresponding to dilution rate of D = 0.09 h^{-1}. These substrate losses can be further reduced to 0.22 g/litre in a fourth stage. Fig. 8 summarizes the performance of a multistage system at a dilution rate of D = 0.09 h^{-1}. It shows that the gain in cell output only increases from 1.31 to 1.33 g/litre h^{-1} in the last stage (Meiering et al. 1977).

The total production of a three-stage system would amount to 78.6 kg in a 20-h fermentation period as compared to 42 kg in the 20-h exponential growth phase in the batch system. The batch process requires several hours of inoculate preparation in a separate fermentor. In addition to this, a lag phase of 6–8 h is encountered after transferring the inoculum to the main fermentor. Taking these delays into account as an additional time of 12 h, a ratio of $[(VX_m)_{\text{batch}} / (DX_m)_{\text{cont. flow}}] = 0.33$ would result from comparison of the production potentials of the two systems. This means that a three-stage continuous flow system, as shown in Fig. 9, could operate with 1000 litre vessel capacity in each stage to produce the same amount of biomass as the 3000 litre batch fermentor. Component design for the individual stages would be facilitated by this reduction factor of approximately 66%. Especially, vibration problems of the fermentor shaft, as illustrated in Fig. 10, for the 3000 litre model could be controlled more readily without the proposed installation of an expensive pressure

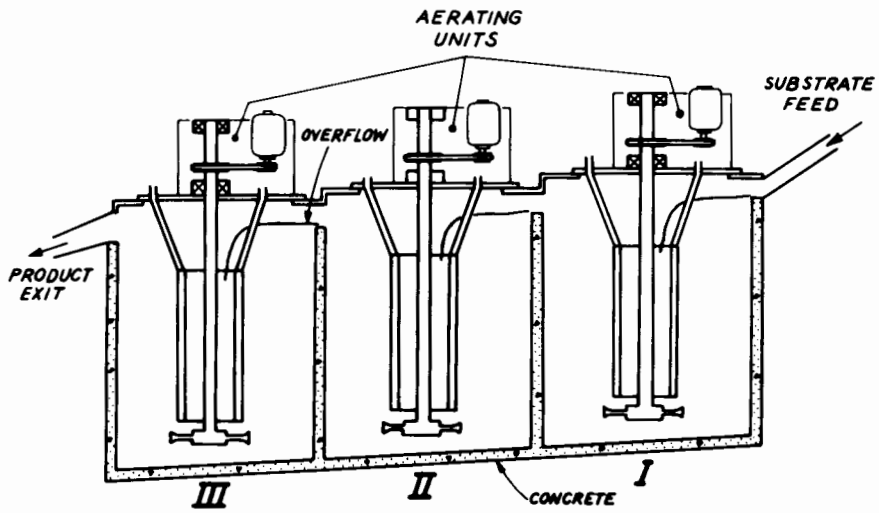

Fig. 9. Layout of 3-stage continuous flow fermentation system with aerating units developed for batch process.

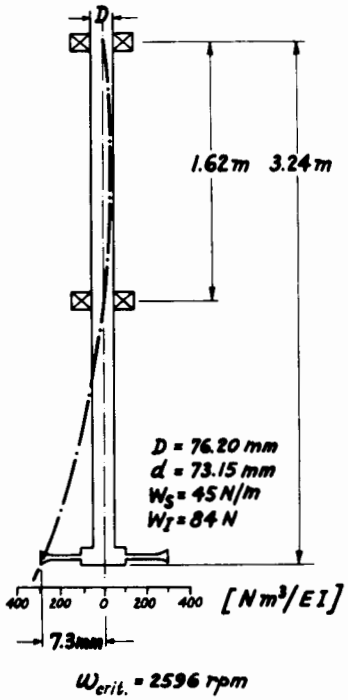

Fig. 10. Shaft deflection in 3000 litre fermentor.

seal and bearing at the vessel bottom. Construction cost could be reduced in both batch and continuous flow systems by using simple containers made of concrete on the farm rather than manufactured stainless steel units. Aerating units of relatively small size could be suspended from a frame construction or the vessel ceiling, as shown in Fig. 9.

Pollution control will likely require a lower carbohydrate content than 1 g/litre in the effluent. The cost of adding a fourth stage in a continuous flow system to satisfy these requirements could render it economically inferior to the batch systems. Another possibility is a progressively higher rotor speed in the sequential stages to improve reaction speed and oxygen supply. Rotor speeds, however, are limited by their shearing action on the microbes. Nevertheless, a limited experiment showed that a fully developed culture can sustain significantly higher speeds than an inoculum in the lag phase (Azi 1976). The behaviour of the culture and the performance of a multistage stage could only be outlined in this report on the basis of batch fermentation data, but should be investigated in an actual pilot scale system.