

PROCESS BIOTECHNOLOGY IMPLICATIONS FOR CHEMICAL ENGINEERING AT THE UNIVERSITY OF THE WEST INDIES

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INTRODUCTION

The International Bioindustry Forum has stated that Biotechnology refers to the application of living organisms and their cellular, sub-cellular or molecular components to create products and processes (1). These techniques can be used to introduce desirable characteristics in existing biological species more rapidly and precisely than has been possible with conventional practices. The aim is to produce processes which provide and encourage environmentally sustainable development. It is hoped that existing products will be produced in greater quantities and the new processes will be more efficient. Process Biotechnology or Biochemical Engineering involves interfacing process engineering principles and practice with the biological sciences. It involves studying the opportunities that the latter offer and impose upon fermentation, fermenter design and control, product separation and materials handling.

As part of its development thrust in the nineties the Department of Chemical Engineering recognised the importance of process biotechnology as an important area for development in the Caribbean region. It supported the view that Biotechnology in its most general sense can help meet societal needs. Internationally, it has demonstrated the potential to satisfy current and future human needs and expectation in human health care, agricultural practice, food and feed supply. The social, commercial, industrial and environmental benefits to this Caribbean region has to be investigated. The Department has been carrying out limited research in the field of Process Biotechnology. Under the Inter-American Development Bank Loan programme, it is establishing laboratory facilities in the area. This is scheduled to include a microbiology laboratory, laboratory scale process biotechnology equipment and small pilot plant facilities. This paper identifies the major work undertaken in the area of Process Biotechnology, possible future trends and limitations associated with the new thrust. The comments are chiefly those of the author.

GENERAL INDUSTRY AND PROCESSES

Bio-process operations may be listed in Table 1 (2). These are the same as in mainstream Chemical Engineering Processes. Research in biochemical engineering is usually based upon the fundamental biochemical and physical aspects of the design of unit operations applicable to product fractionation and recovery, isolation and utilisation of microbes. Work also includes the scale-up applications of immuno-affinity adsorbents in fixed or fluidised beds, the use of affinity HPLC sensors in bio-quantitation, control of production and recovery (3). Additional work is concerned with solid state fermentation, the application of membrane processes for solid-liquid separations, molecular fractionation and solvent extraction using near critical carbon dioxide and foam fractionation processes. Research and development has also been in the areas of microbial proteases from fermentation, pigments from food process plants and functional proteins present in wastes (4).

The Caribbean region has a diverse and suitable supply of tropical fruits, vegetables crops and other agricultural waste products. The major industries worldwide with biotechnology applications may be listed as:

1. Pharmaceutical and Vaccines
2. Food and Feed
3. Pulp and Paper
4. Textiles
5. Leather
6. Biocatalysts
7. Forestry and Agriculture

The major industries in the Caribbean with biochemical engineering applications are covered under 2 and 7. These industries generate waste that can have bio-process applications.

During the processing of raw agricultural produce into consumer products, a number of materials become available in the form as listed (5):

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PROCESS STEPS	UNIT OPERATIONS	
Sterilisation	Thermal Membrane/Chemical	
Fermentation	Mechanical Agitation Immobilised Cell Sedimentation	Packed Bed Fluidised Membrane
Cell Separation	Centrifugation Filtration Sedimentation	Ordinary Micro-filtration
Cell Disruption	Mechanical Enzymatic Chemical	Homogenisation Ball Milling
Product Recovery	Extraction Absorption Adsorption Crystallisation	Precipitation Distillation Centrifugation
Concentration	Reverse Osmosis Evaporation Ultrafiltration	
Purification	Chromatography Electrodialysis Crystallisation	Ion Exchange Gel Permeation HPLC Affinity
Polishing	Diafiltration Drying	Spray Drying Lyophilisation

Source: Brooks (2)

Table 1: Bioprocess Operations

Outer covering	- husk, hull, shell, testa, skin, fibre, bran, linter
Inner Core	- stone, seed, cob
Factory dust	- sawdust and coir dust
Partially Processed Residues	- prawn shell and head, low grade fish, bones, hair

Residues are also available on the farm as straw, stems, stalks, sticks, trash and leaves, dung, urine, litter, poultry and pig excreta. The four categories according to availability may be listed as follows in Table 2.

Conventionally, organic residues are used as soil conditioners, animal feed, litter and fuel. Research and development work has centred on a number of new uses which include the manufacture of:

Chemical and specific products	Furfural, activated carbon alcohol, organic acids, polysaccharide, chitosan wax
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Animal feed	Pretreated and upgraded products, microbial biomass, single cell protein
Food	Microbial biomass, fermented food, beverages, mushrooms, oil and proteins

From this vast array of possibilities the Chemical Engineering Department will have to identify the areas of process biotechnology that would be relevant to the needs of the Caribbean area. Presently in the Caribbean, alcohol, rum, beer, anhydrous alcohol, vinegar and acetic acid are already industrially established. Industries with major export potential are the sugar, citrus, banana, brewing, distillation and food and vegetable processing industries. Research work at the Department of Chemical Engineering has centred around the production of ethanol from various substrates such as cane tops, grasses, fruits and vegetables (6). Work has also been centred on acquiring fundamental knowledge in the production of citric acid, dextran, baker's yeast and vinegar using

molasses as substrates.

The suggested areas for continuing research and development can be listed as follows:

1. Fermentation and Sterilisation
2. Solid-State fermentation
3. Protein Production and recovery from leaves and Carbohydrate wastes
4. Use of Enzymes
5. Isolation and usage of Indigenous microbes
6. Environmental Applications.

Genetic manipulation of microcubes is an area of research that the Department cannot enter at this time with current human and physical resources.

Environmental applications are not covered in this review.

BRIEF OVERVIEW OF RESEARCH - FUTURE TRENDS

Fermentation

(I) Ethanol

The Department has been experimenting with the production of Ethanol from various feedstock associated with the sugar industry and the banana industry such as molasses, ligno-cellulosic wastes, reject syrup and banana pulp. The concept utilised is that of a plant capable of producing ethanol from a

<p>(1) Agro-Industrial</p> <ol style="list-style-type: none"> 1.1 Rice Milling 1.2 Sugar Industry 1.3 Coconut Industry 1.4 Coffee Industry 1.5 Banana Industry 1.6 Fruit & Vegetable 1.7 Poultry Industry <p>(2) Factory Effluents</p> <ol style="list-style-type: none"> 2.1 Distillery Effluent 2.2 Sugar Factory Effluent 2.3 Dairy Effluent 2.4 Fruit & Vegetable 2.5 Abattoir Effluent <p>(3) Crop Residues</p> <ol style="list-style-type: none"> 3.1 Straw/Stalk/Stem/Stick <ul style="list-style-type: none"> Rice Straw Banana Stem Coconut Stem 3.2 Shell/Cob/Husk/Trash <ul style="list-style-type: none"> Groundnut Shell Nutmeg Shell Sugarcane Trash 3.3 Leaves <ul style="list-style-type: none"> Coconut Leaves Cassava Banana 3.4 Animal Wastes <ul style="list-style-type: none"> Dung and Urine Biogas Slurry Poultry Excreta <p>(4) Human Habitation Wastes Solid and Liquid Waste</p>	<p>By-products</p> <ul style="list-style-type: none"> Rice husk, bran Bagasse, molasses, filter press mud, cane trash Husk, pith, shell, water, copra Husks Fibre, reject bananas, stems and leaves Mango waste, citrus waste Feathers, shells
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Table 2: Organic Residues

range of raw materials (7).

Yields of reducing sugars and ethanol have compared favourably with that of other investigators. Some further work is required to improve efficiency and prevent product inhibition (8) (9). McGaw (10) reported that ripe bananas can be regarded as a suitable substrate for ethanol production. He reported that ethanol yield per tonne of bananas may be as high as 100 litres. Fermentation of the "Silk fig" variety of bananas using a two-stage process was reported by Blackwood (11). She obtained a 4.32% w/v acetic acid in three (3) days from a banana liquor containing approximately 9% alcohol by volume. Corn cobs were utilised as packing material.

(II) Citric Acid and other Products

The production of citric acid from sugarcane molasses and juice has been the subject of investigations in the Department (12). Yields so far have been low. This work is continuing, utilising different strains of *Aspergillus Niger*.

A plethora of products of actual or potential importance can be made from sugar by products. These range from detergents to pharmaceutical drugs. Movements into pharmaceutical drugs seem unlikely. The Biochemical Engineering thrust would most likely concentrate on high volume, intermediate value products, such as amino and organic acids and low volume, high value products such as enzymes.

Protein Recovery

Large amounts of nutrients are accumulated and wasted in many processes involving harvesting, handling and processing of fruit and vegetables. Wastage occurs from rotting fruit and reject fruit peels. Processes to recover protein from process waste include:

1. Cell Disruption
2. Protein Coagulation
3. Separation
4. Drying

Work has started on the extraction of protein from the unstrained slurry of fully ripe fermented banana of the "Silk fig" variety. Cell disruption was by sonicator and pressure homogeniser (13). The resulting protein was used to fortify cereal-based, baked products. Results showed that food grade protein can be recovered from fermented banana pulp. Further work is suggested. Initially, this has been expanded to include protein-recovery from rum distillery waste.

Leaf protein can be considered to be a good

potential source of human consumption. The leaves of many crops contain crude protein, ranging as high as 20% in bamboo and lettuce, to above 30% in leaves such as cowpea and cassava (14). Investigations so far have been carried out on protein-recovery from passion fruit skin, cassava, banana and sugar-cane leaves. Yields are promising. Work in process biotechnology would continue this line of research.

Membrane separation techniques can provide high selectivity for removing or concentrating high molecular mass components (2). The use of high-capacity membranes to separate valuable macromolecular components from sugarcane juice during the juice pretreatment stage has been shown to be possible.

Microbiological Implications

The microbiology laboratory should take the lead in identifying indigenous strains of microbes that may be used in any or all of the above processes. *Leuconostoc Mesenteroides* is partly responsible for the staling of cane. This in turn produces dextran which can be a valuable product when purified. Local strains of *Aspergillus Niger* may be identified to assist in making solid state fermentation more efficient. This identification and isolation of useful indigenous microbes should be the aim of the microbiology section.

Solid State Fermentation

This has been attempted on a small scale using *Aspergillus* on wheat bran, bagasse and soya. The work is still in its initial stages. There are two types of processes: one aimed at the total microbial transformation of starch into protein and those aimed at a partial transformation of starch into a protein-enriched product. The process of solid state fermentation consists of fostering the development of selected aerobic micro-organisms on a solid substrate. This results in protein-enriched, fermented foods. This process is based uniquely on filamentous fungi. The Process Biotechnology laboratory is expected to continue this work.

Revilla (15) successfully sequenced the pre-treatment raw cane juice stage using ultra-filtration and electrophoresis to remove water soluble protein. Brooks (2) has stated that such products as waxes and proteins would be rejected by ultra filtration techniques, while nano-filtration procedures could be used to separate sugar from the mother liquor containing bio-products such as carboxylic acids and amino acids. He has suggested that by applying modern

separation techniques, it may be possible to produce a source of renewable by-products for the sugarcane industry. The Process Biotechnology laboratory can undertake this study as part of its research and development.

Enzyme Technology

Enzymes have been utilised for the hydrolysis of lignocellulosic materials and starch-based materials. Work has been reported on the malt hydrolysis of potato and eddoe starches (16). Subsequent work has been on the production of the amylase enzyme from *Bacillus Subtilis*. The strain utilised has been shown to be capable of hydrolysing indigenous starches such as arrowroot starch (17).

It is envisaged that work will continue in this area to product high fructose syrups and ultimately ethanol, if so desired. The amylase produced has application in the sugar industry to remove starches which hinder proper juice clarification.

Work has also been done as a research project together with the Caribbean Industrial Institute on enzymatic treatment of mango waste. The intention was to increase the yield of fruit juice and reduce the viscosity of the juices. Results showed that a satisfactory product can be obtained from this treatment.

CONCLUSION

The paper reviewed the general area of process biotechnology. It reviewed the current and past research in the Department in this area. Suggested areas of research for the new thrust in process biotechnology are in fermentation, enzyme technology and in isolation and identification of local microbes with the potential for product applications.

REFERENCES

1. Publication of the International Bioindustry Forum (1992). "Policies for Sustainable Development." UNCED Washington DC 11 pages.
2. Brooks, S.A. (1993). "A Strategy for the Production of Chemicals from Biomass in the Agro-Industrial Sector of Caribbean Countries". Prepared for Commonwealth Science Council, London 20 pages.
3. Directory of Research (1993) Chemical Engineering Department, University of Birmingham.
4. Morris, G.G., Imrie, F.K.E. and Phillips K.C. (1973). "The Production of Animal Feedstuffs by the Submerged Culture of Fungi on Agricultural Wastes". Proceedings of Fourth International Conference on Global Impacts of Applied Microbiology, Sao Paulo, Brazil, July 23rd - 28th.
5. Vimal, O.P. (1982). "Recycling of Organic Residues". Status and Trends in India UNEP Industry and Environment April/May/June pp. 7 - 8.
6. Mellowes, W.A. and Lewis, R.E.R. (1987). "Potential Sources for Ethanol Production in Trinidad and Tobago". Proceedings Inter-American Sugarcane Seminars, September pp. 250 - 268.
7. McGaw, D.R. (1981). "The Potential for Biomass as an Energy Source for the Caribbean". Proceedings Commonwealth Engineers Conference, Trinidad Vol. 1 pp. 72 -81.
8. Inkim, C.C. and Mellowes, W.A. (1985). "Hydrolysis of Cane Tops and Guinea Grass". Proceedings Sugar Association of Caribbean Conference, March pp. 118 -132.
9. Mellowes, W.A., Inkim, C.C. and Maharaj, D. (1992). "Evaluation of Hydrolysis Methods for Conversion of Selected Tropical Biomass to Fuel Ethanol". Proceedings Energy from Biomass and Wastes, XVI Institute of Gas Technology March.
10. McGaw, D.R., Pilgrim, A.C. and Ameerli, N. (1986). "The Production of Ethanol from Bananas". West Indian Journal of Engineering. Vol. 11(2) pp. 5 - 11.
11. Blackwood, A.R. (1992). "Production of Banana Vinegar". M.Sc Project Report, Food Technology Unit, The University of the West Indies.
12. Mellowes, W.A., Tota-Maharaj B. and Younger E. (1991). "Citric Acid Production from Cane Juice and Molasses". Proceedings West Indies Sugar Technologists, XXIV Conference April pp. 230 - 235.

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13. Zewdie-Bosuner, A. (1994). "*Protein Recovery from Fermented Banana (Musa spp.)*". Pulp. M.Sc Project Report, Food Technology Unit, The University of the West Indies.
14. Berry, R. (1986). "*Tropical Seeds, Fruits and Leaves as sources of Protein Plant Protein*". Edited by Robert L. Ory.
15. Revilla, J.G., Gonzalez, N. Kopecky, J. and Votruba, L. (1976) Revista ICIDCA. Vol. 10(3).
16. Hosein, R., Mellowes, W.A. (1988). "*Malt Hydrolysis of Sweet Potatoes and Eddoes for Ethanol Production*". Biological Wastes 9 pgs.
17. Doodnath, V. (1994). "*Production of Alpha Amylase by Bacterial Fermentation with Bacillus Subtilis*". M.Sc Project Report, Food Technology Unit, The University of the West Indies.