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Review

The status and trends in food, industrial and environmental biotechnology research in Zimbabwe

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Biotechnology offers industrial and environmental solutions through the use of living cells and or their products to provide goods and services. This paper reviews for the first time the status and trends in industrial and environmental biotechnology research in Zimbabwe for the past 25 years. An attempt is made to pinpoint significant research gaps, and the applications of the research in critically reviewing the research. Zimbabwe is pursing an active research and development in food and industrial and environmental biotechnology. Some of the notable successes from these researches are summarised in this review serving as guidance to scientists on what needs to be investigated further, for partnerships and for those who wish to develop the biotechnology to commercial products. A number of enzymes with potential industrial and environmental applications have been isolated from untapped bioresources in Zimbabwe. Some enzymes were purified and characterised and await applications in the industry. Ethanol and biogas which are renewable energies can be produced by converting biomass using microorganisms. Much has been published on functional properties of mucilage and gum from plants in an attempt to facilitate the use of the polymers in industry. There have been some efforts to treat waste (water) using aerobic and anaerobic digestion into biogas and research to degrade dyes from textiles industry using enzymes from novel fungi. So far little has been done to apply much of the research findings in pilot- and large-scale production systems.

Key words: Biotechnology, industrial enzymes, environmental biotechnology, Zimbabwe.

INTRODUCTION

Biotechnology is a broad term that refers to techniques that use biological systems, living organisms, or derivatives thereof, to make or modify products and to provide goods and services to people. It has wide applications in agriculture, medicine, industry, food science and the environment. Biotechnology provides potential to be the source of solutions to many economic, social and environmental problems that developing countries are confronted with. It has the potential to produce new, improved, safer, and less expensive products and processes. Pharmaceuticals and diagnostics for humans and animals, seeds, entire plants and animals, foods and food additives, industrial enzymes, bioenergy and pollution degrading microorganisms are just a few of the things that can be created or enhanced through the use of biotechnology.

Biotechnology uses living cells from plants, animals and microorganisms (yeasts, moulds and bacteria) as well as enzymes from the living cells to produce value added products and services. Living cells can be used as they are or improved to work as “cell factories” to produce enzymes, antibiotics, vitamins, vaccines and proteins for different uses. Biomasses like starch, cellulose, vegetable and agricultural and industrial waste are used in the production of chemicals (green chemicals), biodegradable plastics, biofuels like ethanol, biodiesel and biogas among other things. Successful application of biotechnology integrates a multiple of scientific disciplines such as microbiology, biochemistry, genetics, molecular biology, chemistry and chemical engineering. Biotechnology can reduce pollution and waste, decrease the use fossil fuels, raw materials and water, lead to better quality food products, create new bio-based materials and provide environmentally friendly alternative to some chemical and physical processes. Efficient uses of natural biological resources as a strategy for sustainable industrial growth are paramount in achieving economic, social and environmental sustainable human

development.

The ability of a country (its private firms and public agencies) to research, develop and trade in new biotechnology goods and services defines the national competitiveness of a country in biotechnology. In most countries, researches on biotechnologies related to agriculture and medicine receive more coverage than those related to industry, food and the environment. In recognition of the importance of biotechnology, this paper reviews the status and trend in industrial and environmental biotechnology research in Zimbabwe over the last 25 years. These researches have been largely been done by universities and other research institutions.

FOOD AND INDUSTRIAL BIOTECHNOLOGY

Metabolic reactions are mediated by enzymes which catalyse the reactions with great specificity and rate enhancement. Such catalytic properties together with their environmental compatibility and easy disposal nature provide tremendous opportunities for their Industrial applications. Enzymes are being increasingly being used in industry instead of harsher processes which employ thermal, chemical and mechanical technologies to convert raw materials into products. Enzymes are used to catalyse many industrial processes as diverse as textiles and paper manufacture, food processing, diagnostic kits, washing liquids and medicine. Industrial enzymes can be obtained from various sources which include plants, animals and microorganisms. Nowadays many enzymes are being produced at large-scale from microorganisms because of the many advantages associated with it. Many microorganisms and their enzymes have been discovered by means of extensive screening and are now commonly used in food and industrial applications. This discovery of new microbial enzymes through persistent and extensive screening has provided ways of solving some industrial and environmental problems. In Zimbabwe, there has been a significant research to explore the potential industrial enzymes from various biological and natural resources within the country as discussed below.

A variety of enzymes such as proteases, cellulases, lipases with potential industrial application have been discovered by many researchers in Zimbabwe. Cellulose has a great potential as a source of energy and chemical feedstock. The complete hydrolysis of cellulose requires several cellulase enzymes; an endocellulase that cleaves interior glycosidic bonds randomly, an exocellulase that cleaves glucose from the non-reducing end of the cellulose chain, cellohydrobiase that cleaves cellobiose from the non-reducing end of the cellulose chain and cellobiase that hydrolyse cellubiose into glucose. Cellulases have many potentially useful industrial applications.

A number of cellulase producing *Bacillus* bacteria strains have been isolated from hot springs in Zimbabwe and their enzyme production studied under various culture conditions. Mawadza et al. (1996) reported the production of endo-β-1,4-glucanase by a *Bacillus* strain isolated from a hot spring in Zimbabwe studied in batch culture, chemostat culture, and carbon dioxide-regulated auxostat (CO_2-auxostat). The bacteria produced the enzyme in the presence of excess glucose or sucrose, but not under carbon-limited conditions in chemostat using mineral medium. They also observed a specific growth rate dependent linear increase in enzyme production in glucose excess, nitrogen-limited chemostat cultures. A high specific growth rate of 2.2 h^{-1} and a high rate of enzyme production of 362 nkat (mg dry mass-h)$^{-1}$ were attained under nutrient rich conditions in the CO_2-auxostat. Increasing the yeast extract, tryptone and glucose concentration of the medium increased the specific rate of enzyme production. The *Bacillus* strain HR68 used in this study produced endo-β-1,4 glucanase only and none of the other cellulase enzymes.

Mawadza et al. (2000) further purified and characterised the cellulases produced by two bacillus strains CH43 and HR68 isolated from hot springs in Zimbabwe. The cellulases produced by the two *Bacillus* strains, CH43 and HR68 were purified to homogeneity from culture supernatants. Both enzymes had molecular mass of 40 kDa and isoelectric point of 5.4 and also resembled each other in N-terminal amino acid sequence which was Ala-Gly-Thr-Lys-Pro-Val-Ala-Lys-Asn-Gly-Gln, showing 100% homology with endoglucanases from *Bacillus subtilis*. The cellulases were optimally active in the pH range 5 - 6.5 and had optimum temperature of 65 and 70°C for the endoglucanases from CH43 and HR68, respectively. The enzyme retained complete activity for at least 24 h at 50°C. The enzymes showed highest activity with β-glucan as substrate followed by carboxymethylcellulose. The enzymes did not possess any dramatic characteristics with respect to their activity and stability under extreme conditions. However, in view of their ability to hydrolyse crystalline cellulose rather effectively as compared to the majority of other known endoglucanases, the authors recommended that the enzymes were interesting candidates for further investigations on genetic make-up, structure and understanding the hydrolytic action.

Zvauya and Zvidzai (1995) reported the constitutive production of endoglucanase by a *Bacillus* sp. Isolated from a Zimbabwean hot spring. The sporulating and aerobic *Bacillus* species produced endoglucanase when cultured on medium with initial pH between 5.0 and 9.0 and at 30 to 60°C. Optimal production of endoglucanase was at pH 6.0. The enzyme was constitutively produced when the organism was cultured on starch, cellobiose, carboxymethylcellulose, sucrose, glucose, galactose, Avicel, lactose, mannose or maltose.

Extracellular cellulase production by tropical isolates

of *Aureobasidium pullulans* from Zimbabwe was reported by Kudanga and Mwenje (2005). Isolates were obtained from leaves and fruits of diverse indigenous plants from areas around Bulawayo, Zimbabwe. The isolates were grown in synthetic medium containing cell walls of Msasa tree (*Brachystegia* sp.) as the sole carbon source. All isolates studied produced carboxymethyl cellulase (endoglucanase) and alpha-cellulase (exoglucanase) activity. Endoglucanase-specific activities of ten selected isolates ranged from 2.4 to 12.9 µ mol glucose (mg protein)$^{-1}$h^{-1}, while activities on alpha-cellulose (exoglucanase activity) ranged from 0.3 to 22.4 µ mol glucose (mg protein)$^{-1}$day^{-1}. Carboxymethyl cellulose induced the highest cellulase activity in the selected isolates. The study indicated that some *A. pullulans* isolates of Zimbabwe origin produced significant extracellular cellulolytic activity and that crude cell walls may be good inducers of cellulolytic activity in *A. pullulans.* The existence of β-glucosidase in the same *A. pullulans* isolates by Okagbue et al. (2001) meant that these A. pullulans isolates can produce the complete cellulase activity. However, the extracellular α- and β-glucosidases were generally low under the conditions of cultivation used and addition of Tween 80 to a 24 h old culture led to a three-fold increase in yield of α-glucosidase (approximately 6.0 U/ml).

Cellulolytic and pectinolytic activities of five *Capnodium* isolates (sooty mould) from Zimbabwe were investigated by Mwenje and Mguni (2001). The five isolates showed the ability to produce polygalacturonase, pectin lyase, and endo-1,4-β-glucanase enzymes when cultured in avocado fruits. The results indicated that the *Capnodium* isolates are potential sources of cell wall degrading enzymes, especially pectic and cellulase enzymes.

An endo-polygalacturonase (PGI) from a Zimbabwean species of *Armillaria* was purified and characterised by Mwenje and Ride (1999). The endo-polygalacturonase produced both in vivo in inoculated potato tubers and in vitro on crude cell walls of *Corylus avellana* by an isolate of a Zimbabwean species of *Armillaria* (group III), was purified 18.8-fold, to apparent homogeneity on SDS-PAGE, by gel filtration followed by cation-exchange chromatography. The PGI was highly stable throughout the purification process. Its molecular weight was found to be 40 kDa and the enzyme was not glycosylated. PGI had an isoelectric point of 7.3, showed activity over a broad pH range, with an optimum at 5.5 and showed a preference for polygalacturonic acid to pectin, with a polygalacturonic acid to pectin ratio of 1.4. Although the authors were interested in the role of the PGI in the phytopathogenicity of *Armillaria*, the *Armillaria* isolates can be potential sources of polygalacturonase. Muzariri et al. (2001) screened 224 fungal strains from Zimbabwe for ligninolytic and cellulolytic activities and reported that fungal strains in Zimbabwe represent a potential source of these enzymes.

In Zimbabwe, there has been a great interest in lipases which have potential application in biotechnology from local sources. The areas of application in biotechnology that have been investigated for lipases include ester synthesis, tryglyceride modification, phospholipid modification, monoglyceride production and fatty acid production through triglyceride hydrolysis (Ncube and Read, 1995). Free fatty acids are widely used in industry but are only found in small quantities in nature. A lipase from *Vernonia galamensis* seeds from Chiredzi Agricultural Research Station, Zimbabwe, was evaluated for its potential use in biotechnology by Ncube et al., (1995). An acetone powder was prepared from *Vernonia galamensis* seeds and used as a source of crude immobilised lipase to characterise the *Vernonia* lipase and its potential use in biotechnology. The lipase showed no fatty acid specificity in hydrolysis of coconut and soya bean oils. Short chain triglycerides were the preferred substrates in transesterification reactions. In both transesterification and hydrolysis of 1,3-dipalmytoyl-2-oleylglycerol the lipase showed selectivity for the 1,(3) position of triglycerides. The acetone powder catalysed the hydrolysis of triglycerides in 2,2,4-trimethylpentane (TMP) and was evaluated for use in the continuous production of poly-unsaturated fatty acids from soya bean oil in a packed bed reactor. Complete hydrolysis of 2.5% w/v soya bean oil dissolved in TMP was achieved within 3 h of introduction of the oil solution and the hydrolysis decreased with time depending on temperature conditions. The fact that lipase activity was found in the ungerminated seed and the characteristics showed by the lipase make *Vernonia galamensis* an attractive oilseed crop not only as an industrial oil source but also a source of cheap lipase.

Two new esterases (JEA and JEB) and a lipase were isolated from the seeds of *Jatropha curcas* L. from Zimbabwe by Staubmann et al. (1999). Lipase activity was only found during germination of the seeds and increased to a maximum after 4 days of germination. All enzymes were found to be active in the alkaline range at around pH 8 and the purified esterases were very stable at high temperatures. The molecular weight (SDS-PAGE) of both esterases were determined to be 21.6 - 23.5 kDa (JEA) and 30.2 kDa (JEB) and the isoelectric points were 5.7 - 6.1 and 9.0 for JEA and JEB respectively. Both esterases hydrolysed tributyrin, nitrophenyl esters up to chain length of = C4 and naphtylesters up to chain length of = C6. In transesterification reactions, *Jatropha* lipase was found to be most active at very low water activities (0.2) and in high water activities, the lipase hydrolysed triglycerides into conversions above 80%. The lipase hydrolysed both short chain and long chain triglycerides at about the same rate. *Jatropha* lipase is a potentially useful biocatalyst in the hydrolysis of triglycerides in organic solvents. The esterases and lipase from *Jatropha curcas* in this study showed interesting characteristics such as high stabilities and

activity at low water activity.

Extracellular esterase activities of seven selected *Aureobasidium pullulans* isolates from Zimbabwe were studied under varying conditions using para-nitrophenol acetate as substrate (Kudanga et al., 2007). Esterase activities in broth cultures ranged from 0.011 - 0.223 mmol/µg protein/min while activities ranged from 1.5 - 12.8 U/ml under solid state fermentation. The esterases were optimally active at pH 7.6 - 8.0, showed temperature optima of 35°C and retained more than 50% activity at 60°C and pH 4.0 - 7.0 after 150 min. The *A. pullulans* esterases differed from most fungal esterases in that they were optimally active in alkaline conditions and active over a broad pH range. Since the microorganism is GRAS cleared, it can be used to produce enzymes that are potentially useful in various applications in food, detergent, pharmaceutical and bioorganic transformations.

Ncube et al. (1995) studied the fatty acid selectivity of a lipase purified from *Vernonia galamensis* seed. The substrate specificity of the purified lipase was studied in isooctane. The lipase showed pronounced selectivity for the native triacylglycerol, trevernolin. The rate of hydrolysis of triolein, the corresponding non-epoxy triaacylglycerol, was only 3% of that of trivernolin. In the acidolysis of tricaprylin using a mixture of fatty acids, the *Vernonia* lipase also showed selectivity for vernolic acid. Michaelis-Mentein kinetics of the hydro-lysis of triacylglycerols revealed that the observed high selectivity of the *Vernonia* lipase for trivernolin was mainly due to a higher V_{max} for trivernolin. This novel substrate specificity was an adaptation by the seed lipase to the triacylglycerols of the seed oil that contain up to 80% vernolic acid.

A protease from an alkalophilic *Bacillus subtilis* CHZ1 isolated from a Zimbabwean hot spring was purified and characterised by Zvidzai and Zvauya (2001). The proteolytic enzyme produced by *B. subtilis* CHZ1 was purified using ammonium sulphate precipitation, gel filtration and cationic exchange on S-Sepharose fast flow column chromatography. Protease production was higher when the *Bacillus* strain was cultured in a synthetic medium, M162, supplemented with 0.3% (w/v) organic compared to inorganic nitrogen sources. Enzyme production was growth dependent and production was highest when tryptone was used as the nitrogen source. The purified protease belonged to the serine protease class as showed by its maximum activity at pH 8.0, molecular weight of 35 kDa band (SDS-PAGE) and its full inactivation by PMSF inhibitor. The purified protease produced by *B. subtilis* CHZ1 had characteristics consistent with those of alkaline serine proteases.

A xylanase enzyme produced by a *Bacillus* species isolated from selected indigenous fruits of Zimbabwe was purified and characterised by Chivero et al. (2001). The endo-xylanase enzyme was optimally active at pH 8 and stable over a pH range of 6.0 - 9.0 and its optimum temperature was 60°C. The enzyme was partially purified by ammonium sulphate precipitation and gel filtration chromatography. It had a specific activity of 308 nkat/mg protein. This enzyme could have potential biotechnological applications in pulp and paper and food manufacturing industries due to its high specific activity and alkaline pH optima.

Some properties of the polygalacturonase enzyme from four Zimbabwean wild fruits (*Uapaca kirkiana*, *Zizphus mauritiana*, *Tamarindus indica and Berchemia discolour* ripe fruits) were investigated by Muchuweti et al. (2005). The protein concentrations and activities of the enzyme in the extracts ranged from 0.82 to 1.98 mg/ml and enzyme activities ranged from 2 - 6.5 mmol min^{-1} mg^{-1} in the four fruit extracts. Optimum pH of the enzyme ranged from 4.5 - 5.0 and the optimum temperature ranged from 25 to 37°C. The K_m and V_{max} ranged from 0.12 – 0.25 mg/ml and 0.006 - 0.012 mmol reducing groups/min/mg protein, respectively. Polygacturonase enzymes are used in the food industry, mainly in the manufacture of fruit juices.

There have been a lot of research efforts to understand and find applications for the mucilage that is isolated from ruredzo (*Dicerocaryum zanguebarium*), found in Zimbabwe (Benhura and Mavhudzi, 1996). Ruredzo is a creeping plant that grows widely in sandy soils of Southern Africa and is traditionally used as food and medicine for treating measles and facilitating birth in domestic animals and humans. This plant appears to be a suitable candidate for the commercial exploitation of its mucilage, after the mucilage has been properly characterised. The mucilage from ruredzo is a pectic material with molecular weight of about 500 000 dalton and has been shown to contain 8% uronic acids. The polysaccharide, after reduction of carboxyl groups, was shown to consist of galactose, xylose, arabinose and mannose in the ratio 21:19:12:1 (Benhura and Marume, 1993a). Emulsifying properties of mucilage from ruredzo were studied under various conditions of concentration of mucilage, concentration of salt, and pH by Benhura and Marume (1993b). Emulsification was assessed by diluting samples of emulsions in sodium dodecyl sulphate and measuring absorbance. Emulsification capacity increased with mucilage concentration but was decreased in the presence of NaCl and $CaCl_2$. The optimum pH for the formation of emulsions was about 7.4 with poor emulsification at both higher and lower pHs. The emulsifying property is an important function of gums and mucilage in the food industry where they are used for stabilisation of emulsions, suspension of particulates, control of crystallisation, encapsulation, formation of films and thickening. Crosslinked mucilage prepared from ruredzo was used in the purification of polygalacturonase extracted from ripe tomatoes (Benhura and Mavhudzi, 1996). Used crosslinked mucilage could be regenerated and effectively used for

the purification procedure. Some properties of the ruredzo mucilage crosslinked with epichlorohydrin were reported by Benhura and Mavhudzi-Nyambayo (1997). The depolymerisation properties of mucilage from ruredzo by ascorbic acid in the presence of catalysts were reported by Benhura and Mavhudzi-Nyambayo (1999). No change was observed in the viscosity of samples from ruredzo when treated with 5 mM ascorbic acid. When treated with ascorbic acid in the presence of copper and iron, the amount of reducing sugars increased. The viscosity behaviour of the mucilage extracted from ruredzo was investigated by Benhura and Marume, (2006) after cations from the mucilage were removed by Dowex 50W-X12 resin. Addition of $CaCl_2$, NaCl and sucrose decreased the viscosity of the decationised mucilage studied.

There has been some interest in the polysaccharide extracted from the fruit of *Cordia abyssinica*, a small tree found in Zimbabwe and other parts of southern Africa. The fruit pulp, a sweet mucilaginous flesh that is highly viscous, has been used by rural school children as glue. The fruit is not normally consumed by humans. The polysaccharide of *C. abyssinica*, consists primarily of galactose (27%), rhamnose (21%), mannose (17%), xylose (11%), arabinose (9.5%), glucose (10%), galacturonic acid (5%) and about 2.6% protein (Benhura and Chidewe, 2002). The polysaccharide could not be hydrolysed completely by sulphuric acid under the conditions investigated. The emulsifying properties of the polysaccharide isolated from the fruit of *C. abyssinica*, were investigated under various conditions of concentration of mucilage, concentration of salt, and pH by Benhura and Katayi-Chidewe (2004). Addition of increasing concentrations of the polysaccharide up to 1% enhanced emulsification and emulsion stability. Above 1% concentration the polysaccharide solutions were too viscous for making emulsions conveniently. At a constant concentration of the polysaccharide, addition of salt up to 1% concentration of salt enhanced emulsion formation. The stability of the emulsion formed decreased on increasing the concentration of salt above 1%. Emulsifying ability increased with pH between 7 and 11 but there was a decrease in emulsifying ability above pH 11.

Ethanol production by fermentation of sweet-stem sorghum juice using *Saccharomyces* yeast strains from various sources was investigated by Bulawayo et al. (1996). S. strains Vin 7, SB9, N96 and GSL had more than 85% sugar conversion efficiencies. Sweet-stem sorghum is a potentially good alternative crop to sugar cane for ethanol production. Bvochora et al. (2000) applied very high gravity technology to the co-fermentation of sweet stem sorghum juice and sorghum grain and reported levels of ethanol three times higher than the levels under normal fermentation conditions. The technology involves preparation and fermentation of mashes containing 300 or more grams of dissolved solids per litre compared to 11 - 12% dissolved solids per litre in normal fermentation. This creates an opportunity for process improvements in the conversion of biomass to fuel alcohol which would result in more favourable production economics. An independent and secure source of liquid fuel is seen as a sensible strategy because of Zimbabwe's geographical position, its politically vulnerable situation and foreign-exchange limitations, and for other economic considerations. Zimbabwe has no oil resources and all petroleum products must be imported, accounting for nearly $120 million per annum on average in recent years which amounted to 18 per cent of the country's foreign-exchange earnings. In1980 Zimbabwe pioneered the production of fuel ethanol from sugarcane molasses for blending with gasoline in Africa. Initially a 15% alcohol/gasoline mix was used, but due to increased consumption, the blend was reduced to about 12% alcohol. Annually, production of 40 million litres was possible in 1983 though actual production stood at only 6 million gallons in 2004. However, due to many problems the ethanol production from sugarcane was stopped in the late 1980s but there is now a renewed interest in the project due to high fuel cost and shortage in the country.

ENVIRONMENTAL BIOTECHNOLOGY

The emergence and acceptance of the concept of sustainable development warrants that the scope of environmental biotechnology be enlarged to address issues like environmental monitoring, restoration of environmental quality, resource/residue/waste-recovery/utilisation/treatment, and substitution of the non-renewable resource base with renewable resources. This paper also gives an overview of the research and applications in these subareas of environmental biotechnology in Zimbabwe.

Harare, the capital city of Zimbabwe is facing serious water and wastewater management problems. Harare drains into Lake Chivero while it also abstracts raw water from the same reservoir. Therefore, the lake serves as a sink for pullutants that are not effectively removed via wastewater treatment. Waste-water treatment plants in Harare are overloaded owing to rapid population growth and rapid industry expansion among other factors. Wastewater is believed to be the major direct and indirect source of pollution in Lake Chivero (Moyo, 1997; Nhapi et al., 2001). There is a serious need to research and develop technologies to minimise many of the environmental problems Zimbabwe is facing.

A study of the industrial anaerobic treatment of opaque beer brewery wastewater using a full-scale Upflow Anaerobic Sludge Blanket (UASB) reactor was carried out over a period of two years by Parawira et al. (2005). The anaerobic treatment of the opaque beer wastewater

enabled the brewery to meet the requirements for wastewater discharged into the municipal sewage system of Harare, and therefore the costs of wastewater discharge to the company. The UASB reactor generally reduced the organic load to permissible levels during the period of the study, although there is need to improve its performance in terms of organic load removal. Further benefits from the plant could be realised by taping the energy generated by the anaerobic process in the form of biogas. The methane could be used to heat the steam boiler at the brewery or converted to electricity via a motor generator. The installation of this UASB reactor by the brewery is an attractive economic and environmental alternative considering that we are in era of critical energy shortage, substantially higher energy prices and high demand on environmental protection.

Millions of tons of solid waste are generated each year from municipal, industrial and agricultural sources. Unmanaged organic waste fractions from farming, industry and municipalities decompose in the environment resulting in large scale contamination of land, water and air. The wastes not only represent a threat to environmental quality, but also possess a potential energy value that is not currently being utilised despite the abundance of the solid wastes. These organic waste solid wastes can be treated using anaerobic digestion for biogas production as was investigated by Parawira et al. (2004), Parawira, (2004), Parawira et al. (2005), Parawira et al. (2006), Parawira et al. (2007), Parawira et al. (2008), Misi and Forster, (2001a,b). These studies were concerned with some important aspects of anaerobic digestion of agricultural, food and industrial organic wastes digested singly or in co-digestion. Studies were performed using batch, one-stage and two-stage processes using laboratory-, pilot-, and full-scale anaerobic bioreactors. Anaerobic digestion is a complex biochemical process carried out in a number of steps by several types of microorganisms in the absence of oxygen. Methane and carbon dioxide (biogas) are the principal end products with minor quantities of nitrogen, hydrogen and hydrogen sulphide. Anaerobic digestion has many advantages over corresponding aerobic processes (Parawira, 2004). For improved understanding of the anaerobic digestion, some of the aspects investigated were the profiles of the hydrolytic enzymes and production of volatile fatty acids during the hydrolysis/acidification stage, and methane yield under various reactor configurations and reactor conditions. The results from these studies demonstrated that agricultural and industrial waste and wastewater are potential substrates for anaerobic digestion for the production of biogas and environmental protection.

The large quantities of wastewater (containing high organic load) from industries could also be treated using aerobic mesophilic or thermophilic treatment as was demonstrated by Zvauya et al. (1994) where aerobic thermophilic treatment managed to reduce the organic load by 67% BOD5 and 75% (total solids). Manhokwe (2007) evaluated the biological treatment methods for potato wastewater produced by a potato processing plant in Harare. Anaerobic treatment was found to enable the plant to meet the required effluent quality for discharge into municipal sewage system.

The effectiveness of biological nutrient removal from wastewater at Hatcliffe Sewage Treatment Works in Zimbabwe using an activated sludge plant was reported by Makaya et al. (2007). The mean effluent quality was within the acceptable standards of Zimbabwe National Water Authority for a period of seven months.

Duckweed systems are a form of natural wastewater treatment technology that is ideal for developing countries because it is cheap, less sophisticated and is easy to maintain and thus has potential in Zimbabwe (Nhapi, 2004). A study was carried out in Zimbabwe to evaluate the performance of duckweed ponds as an option for treating and reusing wastewater in small, decentralised communities by Nhapi et al. (2003). The study focussed on nitrogen and phosphorus removal, operational problems and duckweed application using two full-scale trial plants over a one year period. Moderate nutrient removals were reported in the Nemanwa plant and while the Gutu plant did not perform well. The performance of the two plants suffered from poor design especially in pond depth and short-circuiting. The duckweed later died off due to excessive ammonia. However, with appropriate design the duckweed systems could offer an alternative treatment of community wastewater if properly designed ponds are used. Unfortunately there has never been a follow-up to this study.

Wastewater from textile, paper and printing industries and dye houses is characterised by high chemical and biological oxygen demands (COD and BOD), suspended solids and intense colour due to excessive use of synthetic dyes. Direct discharge of these effluents into municipal plants and/or environment may cause many serious problems. Growth, dye degradation and ligninolytic activity using Zimbabwean white rot fungi were studied with respect to growth temperature optima and dye decolouration by Tekere et al. (2001a). Temperature optima were found to vary from 25 - 37°C amongst the isolates. No lignin peroxidise was detected in any of the isolates but all isolates showed manganase peroxidise and laccase activities. Time related decolourisation studies and optimum pH determinations for the dye Poly R478 degradation by the isolates and significant decolouration rates were noted with the following isolates: *Trametes cingulata*, *Trametes versicolor*, *Trametes pocas*, DSPM95 (species to be identified), *Datronia concentrica* and *Pycnoporus sanguineus*. Tropical white rot fungi species which are widely represented in the hardwood forests of Zimbabwe are the least studied with respect to their biodegradative capabilities (Mswaka and Magan, 1998; Mswaka, 1991).

Ligninolytic enzyme production in selected sub-tropical white rot fungi under different culture conditions was further investigated by Tekere et al. (2001b). Manganese peroxidase and laccase activities were monitored when the isolates were grown under varying concentrations of carbon, nitrogen and Mn^{2+}. *Trametes* species, *T. cingulata, T. elegans* and *T. pocas* produced the highest manganese peroxidase activities in a medium containing high carbon and low nitrogen conditions. High nitrogen conditions favoured high manganese peroxidase activity DSPM95, *L. velutinus* and *Irpex* species. Laccase production by the isolates was highest under conditions of high nitrogen and those conditions with both nitrogen and carbon at high concentrations. Mn^{2+} concentrations between 11 - 25 ppm gave the highest manganase peroxidase activity while laccase activity was influenced by Mn^{2+} levels. The subtropical isolates presented in these studies are promising sources of manganese peroxidase and laccase enzymes which have wide applications.

Laccase production by *Trametes modesta* isolated from Zimbabwe (Mswaka and Magan, 1998) was successfully optimised using a mathematical model (central composite design) by Nyanhongo et al. (2002a). Wheat bran, yeast extract and incubation temperature appeared to be the main factors which influence laccase production by *T. modesta*. Laccase production by *T. modesta* could be significantly enhanced by including veratryl alcohol, copper sulphate or 2,5-xylidine as inducers. The absence of xylanases and the presence of trace amounts of cellulases and mannose activities in contrast to high laccase activity in the crude extract make *T. modesta* highly attractive for application in textile and pulp and paper industries. Nyanhongo et al. (2002b) evaluated the decolourisation of synthetic textile dyes by lacasses from a newly isolated strain of *T. modesta*. All the tested synthetic dyes (anthraquinone, azo, indgo, and triarylmethane) were completely decolourised by *T. modesta* laccase under acidic conditions (pH 3-6). The decolourisation rate of this laccase increased with the rise in temperature to 60°C.

Muzariri et al. (2002) studied the decolourisation of pulp and paper effluent water and the simultaneous production of ligninolytic enzymes by wood-rot fungi isolated from Zimbabwe. The isolated fungi produced lignin peroxidase and laccase which decolourised the pulp and paper wastewater effluent. However, laboratory scale development of the commercially promising fungal isolates and subsequent optimisation experiments were not done. The research was not then implemented by the potential beneficial textile industries. Polycylic aromatic hydrocarbons (PAHs) are widespread environmental pollutants with long residence time in environment and many of them or their metabolites are carcinogenic, mutagenic or both. The PAH biodegradative capacities of four selected sub-tropical white rot fungi; *Trametes cingulata*, *Trametes pocas*, *Trametes versicolor* and DSPM95 were evaluated by Tekere et al. (2005). Both static shallow cultures and extracellular fluids were studied using media contaminated with a defined mixture of PAHs. All isolates were competitive for use in PAH bioremediation. These isolates were found to be capable of degrading an organochlorine pesticide, lindane in batch and packed bed bioreactor system (Tekere et al., 2001c).

CONCLUSION

This paper has summarised most of the research and applications of industrial and environmental biotechnology and shows that research on industrial and environmental biotechnology in Zimbabwe has come of age. This work attempted to put together most of biotechnological researches in peer reviewed scientific journals that have been done in Zimbabwe (or elsewhere by Zimbabweans together with their collaborators) over a period of 25 years for ease of reference and fostering of closer research collaborations. Substantial efforts have been done in the field of industrial and environmental biotechnology through isolation of enzymes and carbohydrates polymers from local bioresources. Bioenergy in the form of ethanol and biogas potential can be realised in Zimbabwe from agricultural, municipal and industrial wastes. There are a lot of untapped areas of biotechnology in Zimbabwe which needs to be developed to commercial products.

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