



Blueprint for Sustainable Management and Utilization of Nigeria's Bioresources

Edited by

**Christie Oby Onyia, B. O. Solomon,
A. M. A. Imevbore & O. T. Ogundipe**



Federal Ministry of Science & Technology

Blueprint for Sustainable Management and Utilization of Nigeria's Bioresources

The mono-cultural nature of the Nigerian economy which is about ninety-five per cent dependent on crude oil, has blinded our planners to the richness of the country in bioresources. This is in spite of the fact that over seventy per cent of the Nigerian populace derive their livelihood from subsistence farming while most of the rural poor depend on wild species of biodiversity.

This blueprint therefore seeks to define a roadmap for the conservation, management and utilization of Nigeria's bioresources such that it can combine her profile's current developmental needs and aspirations, even as it upholds the country's biodiversity and her ecological environment.

It will no doubt provide a useful guide to conservationists, manufacturers, researchers, bio-pharmacists, the academia and students of environmental sciences and related disciplines.

Project Co-ordinator

Christie Oby Onyia, Ph.D.
Director, Environmental Biotechnology
and Bioconservation Department
National Biotechnology Development Agency
Abuja, Nigeria.



Concept Publications

BIORESOURCES AND ENVIRONMENTAL STUDIES

ISBN 978-8406-29-7



Chapter 4

Sustainable Bioresources Management and Biotechnology Intervention

by

Onyia, O. C.¹; Solomon, B. O.¹;
Okafor, Nduha²; Okpokwasili, G. O.²;
Sadiq, H. Y.³; and Ebagbe, R.⁴

4.1 Introduction

THE level of capabilities in the science and biotechnology components of biodiversity and bioresources development has been used as a measure of the capability of a country to identify, conserve, utilize and commercialize her bioresources in a sustainable manner. The countries of the world have been classified into four categories based on the richness of their biodiversity and the level of application of biotechnology for its sustainable management as follows:

- (i) bioresources-poor and biotechnology-poor;
- (ii) bioresources-poor but biotechnology-rich;
- (iii) bioresources-rich but biotechnology-poor; and
- (iv) bioresources-rich and biotechnology-rich.

Some countries in the Middle East, including Saudi Arabia, belong to the first group, many advanced countries like the USA, Germany, France, UK, etc., belong to the second group, while many of the southern countries, like Indonesia, Brazil,

¹ National Biotechnology Development Agency (NABDA), Abuja

² Nnandi Azikiwe University, Awka (Professor Emeritus)

³ University of Port Harcourt, Port Harcourt.

⁴ Federal Ministry of Environment, Housing and Urban Development

Mexico, Nigeria, Ghana, among others, belong to the third group. At the time of this classification, no country belonged to the fourth group (Khoshoo, 1996), which meant that no country in the world was both bioresources-rich and biotechnology-rich. More than ten years after, a few countries that originally belonged to the third group have demonstrated such biotechnological capabilities to prove that a country can simultaneously be rich in bioresources and in biotechnology as a tool for development. Such countries include China, Singapore, Cuba, South Korea, India, etc. Brazil, Argentina and South Africa are fast emerging in this field.

There has been a rapid flow of bioresources from the South to the North (i.e. from the third group to the second group). Even though some emerging developing countries of the South have made great advances in the application of biotechnology for solving some of their national problems, the rate of flow of biotechnology from the North to the South is still not commensurate. The countries of the South need to accelerate their rate of adoption of biotechnology to maintain a sustainable ecosystem. African countries are the worst hit. The difference between biological diversity and biological resources depends on what man does with the natural biological endowments bestowed on him in a given ecosystem. There have been various definitions of biodiversity and bioresources as described below.

4.1.1 Biodiversity

Biodiversity is the sum total of plants, animals and micro-organisms existing as an interacting system in a given habitat. It is therefore the totality of genes, species and ecosystem in a region.

At the global level, it is estimated that there are around 17,980,000 species, but a working figure of about 12,250,000 species. These species exist on land, free state with other organisms. It has been that only about 1,604,000 species of plants, animals and micro-organisms have been described so far (WCMC, 1992) and that fewer than 1% of the total biodiversity of the world have been studied for their ecological relationships and role in the ecosystems (Singh, 2004).

4.1.2 Global Distribution of Biodiversity

Nature has been found to be very benevolent to the developing countries in the distribution of biodiversity on the earth. Tropical regions, which harbour most of the developing countries, contain more than half of the world's biodiversity; they harbour at least 50-70% of the total biodiversity of the world. The distribution is maximum in the equatorial, tropical and subtropical regions, but progressively decreases as one goes to the Polar Regions.

The above picture may be deceitful, with the advances in biotechnology, which has led to the establishment of genebanks. While it is a fact that all genetic resources originally came from *in situ* conditions, the situation has quite changed; much of that diversity can no longer be found "in the field", a great deal of diversity can now be accessed from genebanks as evidenced in Table 4.1:

Table 4.1: Genebanks and Accession in ExSitu Collections, by Region

Region	No of Accession	% Total	Genebanks	% Total
Africa	353,523	6	124	10
Latin America & the Caribbeans	642,405	12	227	17
North America	672,061	14	101	8
Asia	1,533,979	28	293	22
Europe	1,934,574	35	496	38
Near East	327,963	6	67	5
Total	5,464,505	101	1308	100
CGIAR	593,191		12	

Source: FAO, 1998

Crop experts believe that much of the existing diversity – particularly of major crops – has already been collected and is in genebanks.

4.1.3 Biological Resources: Definition

Biological resources, commonly referred to as bioresources, is the scientific terminology for the natural biological wealth that influences human life and well-being. Bioresources has therefore been defined as the living entities which include genetic resources, organisms (both macro and micro) or parts thereof, populations, or any other biotic component of the ecosystems with actual or potential uses or values to mankind. They are important components for the progress and economic activities of a nation. Management of a nation's bioresources must be done in a systematic, sustainable manner so as not to endanger the resources of the future generation

4.1.4 Bioresources from the Wild

Man, for many centuries, has secured the various resources he needed from the diversity of the living world. From the Stone Age era, he has developed tremendous efforts to master the surrounding natural environment, to grow better crops and to breed specific animals. Primitive people encountered an array of potential food sources in their environment before agriculture. There were over two hundred thousand species of flowering plants alone from which to choose, thousands of fresh fruits, nuts, vegetables and grains, although some of them could barely be recognized (Fowler and Mooney, 1990) as modern agriculture has transformed most species both in quality and yield. From both wild and domesticated components of bioresources, humanity derives all of its food, energy, clothing, medicine and construction materials, while the idea that natural resources were unlimited was generally accepted until early 20th century. This belief justified the unchecked systematic development of human populations and activities (Charrier *et al.*, 1996). A change occurred when it was realized that the assumption was not true forever. It has now been established that human demographic expansion both in terms

of space and bioresources has limits. Present human activities have therefore become a threat to the very existence of other living species and possibly of man himself. Economic development processes, particularly in the early stages, tended to rely primarily on the extraction of natural resources.

In the past when human population density was low and people had only limited technologies for resource acquisition and environmental change, humans lived in close harmony with nature and the availability of existing bioresources at all times. Increasing population and development of technologies have resulted in the over-exploitation of bioresources with consequent environmental destruction of income generation resulting in inevitable deterioration of the human life-supporting system. It has been estimated that economic benefits from wild species alone make up to 4.5 % of the Gross Domestic Product (GDP) of the United States of America, worth \$87 billion annually in the late 1970s. Fisheries, mainly from wild species, contributed about 100 million tons of food worldwide in 1989. Wildlife species are dietary mainstays in much of the world. The genes from wild ancestors of crops endemic to developing countries have made a distinctive contribution in crop improvement with considerable gain to the growers (Khoshoo, 1996); thereby making a positive difference both in social and economic terms.

It is imperative therefore, that adequate measures be adopted for the efficient utilization of those abundant resources in our oceans, rivers, lakes and forests, including geological resources. The relationship between society and bioresources needs to be reappraised for deeper knowledge and better sustainable management of the biological resources.

4.1.5 Conservation of Bioresources

Bioresources is an important and irreplaceable biological resource because its extinction is for ever. The strength of most of the developing countries lies in the richness of their bioresources. It is therefore imperative that the conservation of these resources and their sustainable management/utilization have to be central to all developmental planning in these countries. This is because their economy is dependent on

agriculture, horticulture, animal husbandry, fisheries, forestry, medicinal and bioindustrial products, and others. The conservation of bioresources encompasses richness of species in biological communities, processes whereby species interact with one another and with physical attributes within ecological systems, and the abundance of species, communities, and ecosystems on large geographical scales (Harrington *et al.*, 1990). Conservation has ceased to be a function of building a fence round an area, but involves considerable upstream science and technology, including biotechnology. The differences in the bioresources distribution, conservation and utilization between the developing and industrial countries, have been documented by Khoshoo (1996). Their strengths and weaknesses are highlighted in Table 4.2:

Table 4.2: Conservation of Bioresources: Strengths and Weaknesses of Developing and Industrial Countries

Developing Countries	Industrial Countries
♦ Bioresources-rich	- Relatively poor
♦ Backed by indigenous people, local technical knowledge and indigenous systems of medicine	- Largely non-existent
♦ Bioresources supported by cultural diversity	- Largely absent
♦ Genetics, breeding and biotech base poor	- Rich base
♦ Largely <i>in situ</i> conservation	- Largely <i>ex situ</i> , but <i>in situ</i> for their own agricultural biodiversity
♦ Conservation not entirely science-based	- Largely science-based
♦ Largely subsistence or intensive agriculture	- Largely industrial agriculture
♦ Sustainable utilization of bioresources: not possible without capacity building.	- Capacity in existence

♦ Research and development, education and training, and demonstration and extension need enhancement.	- Rich base
♦ Poverty	- Affluence
♦ Largely bioindustrial development	- Largely industrial development
♦ Bioresources-rich/Technology-poor	- Bioresources-poor/Technology

Source: Castri and Younnès (1996) (Adapted).

It is unfortunate that even though developing countries in the tropics are the repositories of agri-bioresources, they have remained the areas of low productivity and high population density. Many of them, particularly those in Sub-Saharan Africa, fall within the 'hunger belt' of the world and are therefore lagging behind in achieving the millennium development goals for food security and poverty reduction. Some of the reasons for these conditions can be found in Table 4.2 and include poor scientific and technological application to conservation, utilization and general management of bioresources. As has been highlighted by Okigbo in Chapter 1, research and development, education and training, demonstration and extension, are necessary ingredients for sustainable management of any nation's natural resources, including the bio-based resources.

In Nigeria, a number of government agencies, national and international non-governmental organizations (NGOs) involved with biodiversity and bioresources, have programmes for conservation, collection, monitoring, training, etc., but in actual practice nothing tangible can be seen on the ground. In view of the fact that developing countries are major *in situ* repositories of bioresources, definite steps need to be taken in these countries, Nigeria inclusive, to save bioresources for humanity at large.

4.1.6 Need for Conservation of Nigeria's Bioresources

The global biosphere can survive when resource utilization is about 1 per cent; anything up to 10 per cent leads to over-exploitation and therefore poses a threat. Twelve per cent of

the global vascular plants are threatened with extinction. Over 5,000 animal species are threatened worldwide. Nigeria's bioresources are no exception; they are under threat as has been discussed in Chapters 1 and 3 of this blueprint. A national survey to determine the *sources* of threat and the *scale* of the depletion and/or destruction and extinction is desirable. The National Biotechnology Development Agency (NABDA), in collaboration with UNESCO, started a national survey in 2006 of the endangered wildlife of Nigeria, targeting major national parks and games reserves. Report of the survey so far reveals that a number of games are endangered. The Nigerian Natural Medicine Development Agency (NNMDA) has been in the forefront of inventorying the country's plant bioresources for their medicinal and aromatic values. There are great potentials in this sector, but signs of over-exploitation by traditional medicine practitioners have already been detected, even though only two products have been developed at commercial level (Nicosan & Ciklavit). A number of Nigerian-based non-governmental organizations like Bioresources Development and Conservation Programme (BDCP), Nigerian Wetland Development Centre (NDWC), etc, have consistently championed projects geared towards conservation and product development. BDCP has gone further to develop a database of medicinal plants of Nigeria. More of such surveys and efforts should be encouraged. Focused research is needed on forest (which is fast being depleted through human activities), desertification and restoration ecology, bioremediation, and marine resources. Marine resources provide many goods and benefits, including bioactive materials, drugs, and food items, and should be characterized and conserved. Biotechnology has become a major tool in conservation biology.

Conservation efforts are associated with scientific and technical problems which are wide-ranging. Sound biosystematic and experimental evolutionary, genetic and breeding studies are needed to work out the details of distributional patterns of life forms, regenerative capacity, population genetics, evolutionary biology, breeding systems, size and extent of gene exchange within the gene pools, etc. Expertise would

be needed in several relevant and cognate fields such as meteorology, land and water management, landscape and restoration ecology, forestry, agriculture and economics. Also worthy of attention is inventorying of a priority list of biota, monitoring their populations and genetic evolutionary effects of population changes together with remedial, rehabilitative and restocking measures, dynamics of agroecosystems, ethnobotany or ethnobiology, as the case may be, and relationship between biological and cultural diversities and traditional methods for their conservation. Botanical gardens, herbaria and genebanks should be developed to aid conservation. It is equally important to build human resource capacity/expertise in biotechnology which is a knowledge-intensive area to employ molecular approaches, including DNA fingerprinting for plant conservation. The Consortium on Barcode of Life (CBOL) is doing a lot in this area and is promising to be of great assistance to the country through NABDA.

4.2 Biotechnology as a Tool for Sustainable Exploitation and Utilization of Bioresources

Biotechnology as a tool has been successfully employed worldwide not only for effective conservation (*in situ* and *ex situ*) but also for sustainable exploitation of bioresources that would lead to product development and commercialization in the following sectors:

4.2.1 Environment

The major environmental challenges in Nigeria have been identified as deforestation, drought and desertification, erosion, floods, land degradation and pollution, solid and liquid municipal wastes disposal and general risks in the industrial and oil sectors, including pesticides and oil spill pollution.

Environmental protection and conservation has always been a special area of global concern amongst the scientific community. This concern engendered the need for a policy of sustainable development in harmony with the environment. The Stockholm Conference in 1972, and the UNCED Conference in Rio de Janeiro in 1992, both focused world attention on areas

of pollution, biodiversity conservation, and sustainable development. Biotechnological approach to pollution control using biological methods is becoming a more acceptable option to other control methods such as chemical methods. Waste recycling technologies that are cost-effective and environmentally safe, are being generated. The use of plants and microbes (phytoremediation/bioremediation) has been found to be very efficient in restoring or remediating polluted sites. Major breakthroughs have been made in these areas:

Bioremediation: Technology for the bioremediation of Manganese and Zinc mine dumps has been developed through integrated biotechnological approach. Oil spill is a major source of land and water pollution in Nigeria, especially in the Niger Delta area; eco-restoration can be achieved through the use of an efficient crude oil and oil sludge degrading bacterial consortium, the so called 'Oil Zapper' technologies. Expertise in this area exists in Nigeria.

Phytoremediation: Phytoremediation of dye and tannery effluents has been developed and standardized. Plants have also been used to remove the high levels of explosives found in the soil in a process known as phytoremediation. Although it has been known that some microbes can denitrify the nitrate explosives in the laboratory, they could not be effective on site due to some unfavourable environmental conditions. French *et al.* (1999) have transferred this degradative ability from the microbe to tobacco plants, and these have produced a microbial enzyme capable of removing the nitrates.

Bioleaching: Several microbial strains have been isolated for leaching of metal ores of Copper, Zinc and Gold. Microbial polysaccharide gellan and xanthan have been produced in high concentration using *Sphingomonas sp.* Production of cyclodextrin has been standardized using microbial strains of *Bacillus firmes* and *Klebsiella pneumoniae*.

New developments such as bioindicators, biosensors, and rapid identification of species diversity using molecular techniques and isolation of microbial consortia are priority research areas. Developing a more biologically oriented

approach towards pollution control would be **extremely important**. Cleaning up the large river systems and **ensuring the destruction of pesticide residue in large slums in the city** are priorities in which a biotechnological approach would be environmentally safe.

4.2.2 Bioresources Utilization in the Oil Industry

While the activities of the oil industry have indeed generated immense financial benefits, they have no doubt also been increasingly attended by large-scale pollution of the environment. In Nigeria, for instance, the highest increases of oil spills occurs in the mangrove swamp zones and near offshore areas of the Niger Delta; these areas constitute the most sensitive and productive ecological areas. Currently, physical and chemical methods have been the most widely used procedures employed towards minimizing the effects of oil spills. Often, these methods are grossly inadequate and ineffective, and may even result in further contamination of the environment. This therefore creates the need for the development and application of better and more effective methods.

Bioresources, for the most part, play a useful role and are commonly utilized as post-impact remedial agents in the oil industry. The biological activities of living organisms usually involve the uptake of organic and inorganic materials from the environment for growth and sustenance of life. By its very nature, petroleum is amenable to biotreatment and, among the many techniques employed to decontaminate affected sites, bioremediation remains top choice (Okpokwasili, 2002 & Solomon, *et al*, 1986). Bioremediation is a general term used to describe the destruction of contaminants by biological mechanisms, including micro-organisms (e.g. yeasts, fungi, or bacteria), in contaminated soil and water. These organisms eat and digest organic substances for nutrients and energy. This technology has advantages such as cost-effectiveness and the potential to remediate an environment without causing any environmental damage (Okpokwasili, 1994). The absence of any engineered organisms, in most cases, makes this technology suitable for many developing countries (Okpokwasili and Ibe, 1998).

(a) Biotreatment of Petroleum Pollutants

The chemical composition of crude oil and even of refined petroleum products includes hundreds of different alkanes and aromatic hydrocarbons. Microorganisms, with the ability to degrade a wide range of these crude oil components, exist ubiquitously as bioresources' agent in the environment and do appear to respond quite rapidly to the presence of petroleum (Lee & Levy, 1991; Layokun *et al.*, 1987 and Solomon *et al.*, 1986). Many bacteria, yeasts and filamentous fungi have been shown to be able to oxidize and assimilate hydrocarbon components. They include species of *Corynebacterium*, *Pseudomonas*, *Micrococcus*, *Acinetobacter*, *Flavobacterium*, *Arthrobacter*, *Bacillus* and *Staphylococcus* (Okpokwasili and Nnubia, 1999; Nweke and Okpokwasili, 2003). The most important yeasts and fungi include *Candida*, *Rhodotorula*, *Sporobolomyces*, *Penicillium*, *Cunninghamella*, *Verticillium*, *Cladosporium*, etc. A palmwine yeast isolate, *Schizosaccharomyces pombe*, has also been found to degrade petroleum hydrocarbons (Amanchukwu *et al.*, 1988, 1989). Some cyanobacteria and algae are also capable of petroleum degradation (Foght *et al.*, 1987). Microbial communities within contaminated ecosystems tend to be dominated by those organisms capable of utilizing and/or surviving toxic contamination (Okpokwasili, 1994, 1998). As a result, these communities are typically less diverse than those in unstressed systems, although the diversity may be influenced by the complexity of chemical mixtures present and the length of time the population has been exposed (McNaughton *et al.*, 1999).

Local Strains of *Pseudomonas aeruginosa* and *Pseudomonas fluorescence* have been reported to have high potential for bioremediation of crude oil polluted site, Petroleum refining and Petrochemical waste waters (Solomon *et al.*, 1986; Layokun *et al.*, 1987; Oboirien *et al.*, 2005 and Ojumu *et al.*, 2005). The refining processes of petroleum lead to the formation of phenol which at high concentration is very toxic. Chlorinated phenols, which are also commonly used, are difficult to degrade and are classified as xenobiotics. This group of compounds require special attention. Normal alkanes in the range C₁₀ to C₂₆ are viewed as the most readily degraded; but low-molecular weight aromatics such as

benzene, toluene and xylene, which are among the toxic compounds found in petroleum, are also readily biodegraded by many micro-organisms. More complex structures (for example, those with methyl branched substituents or condensed aromatic rings), are more resistant to biodegradation. A single bacterial species has only limited capacity to degrade all the fractions of hydrocarbons present and so these organisms work as a consortium.

Bioremediation can take place under aerobic or anaerobic conditions. Under aerobic conditions, micro-organisms consume atmospheric oxygen in order to function. Under anaerobic conditions, no oxygen is present; rather, the organisms break down chemical compounds in the environment to release the energy they need. Sometimes, intermediate products are formed as the biological processes break down the original contaminants. The intermediate products may be less, equally, or more toxic than the original contaminants. Both *in situ* and *ex situ* bioremediation processes have been developed. *In situ* bioremediation treats the contaminated water or soil where it is found. *Ex situ* processes involve removing the contaminated soil or water to another location before treatment. Enhanced bioremediation involves the addition of micro-organisms (bio-augmentation) or nutrients like oxygen, nitrates (biostimulation), to the environment to accelerate the natural biodegradation process. Various applications of microbial bioremediation technology include:

(b) *Bioremediation of Oil-contaminated soil*

- (i) **Land Farming:** This is a solid phase treatment process that has been used for remediating contaminated soils or other wastes. The process involves excavating contaminated soil and placing it on an impermeable surface where it is spread out in relatively thin lifts of approximately 0.3 – 0.6m in thickness. The soil is then periodically tilled using conventional farm equipment (e.g. disc harrow) in order to aerate the soil and promote volatilization. Typically, the soil is left uncovered and is periodically dosed with liquid nutrients. This aerobic *ex situ* process usually involves the use of soil as an inoculum and the supporting medium for microbial growth (Environment Canada, 1995).

(ii) Soil Vapour Extraction: This process encourages the volatilization of contaminants adhering to soil particles while fresh oxygenated air is introduced into the zone of soil contamination. The fresh air is essential in providing a source of oxygen for the indigenous hydrocarbon-degrading bacteria and thus, natural degradation is promoted. The cost for this treatment process is reactively low and it is typically used to treat soils under roadways, buildings and other structures. Limitations would include the high dependency of this treatment process on the air permeability of the impacted soils and the degree of homogeneity (USEPA, 1991, 1992).

(iii) Bioreactor: The use of a bioreactor for remediation of petroleum-contaminated soils is a batch-treatment technique in which the contaminated soil is excavated, mixed with water (and other additives) and treated in reactor vessels (Blackburn and Hafker, 1993). The slurry is mechanically agitated in the reactor vessel to keep the solids suspended and maintain the appropriate environmental conditions. Micro-organisms may be added initially to seed the bioreactor or added continuously to maintain the correct concentration of biomass. Nutrients are added, pH adjustment is practised and temperature may be optimized in the reactor vessel (Nkala, 2000). This system maintains intimate mixing and contact of micro-organisms with the hazardous compounds and creates the appropriate environmental conditions for optimizing microbial degradation of target contaminants (Choi *et al.*, 1999). Residence time in bioslurry reactors varies, depending on the nature of the soil or sludge matrix, the physical/chemical nature of the contaminant, including concentration, the biodegradability of the contaminants and the desired levels of removal. Residence times of 5 days for PCP-contaminated soil, 13 days for a pesticide-contaminated soil and 60 days for refinery sludge, have been reported (Holden *et al.*, 1997).

Okpokwasili and Oton (2004) have reported a residence time of 28 days for petroleum refinery sludge. Aside from the biodegradability of a particular compound, other limiting factors include the presence of inhibiting compounds

(like heavy metals and chlorides) and operating temperature. The operating temperature range is approximately 15 – 70°C. Dissolved oxygen is also critical and must be monitored along with pH, nutrients and waste solubility. Upon completion of the treatment, the water is removed from the solids which are disposed of or treated further if they still contain pollutants. Dewatering devices that may be used include clarifiers, pressure filters, vacuum filters, sand drying beds or centrifuges. Depending on the nature and concentration of the contaminants and the location of the site, any emissions may be released to the atmosphere, or treated to prevent emissions. Fugitive emissions of volatile organic compounds, for instance, can be controlled by modifying the slurry-phase bioreactor so that it is completely enclosed (Holden *et al.*, 1997). Nwokoro and Okpokwasili (2003) have used the bioreactor system for treating oil-contaminated Niger Delta sediments with success.

(c) Bioremediation of Oil-contaminated Water

Water is a more sensitive medium than soil and requires somewhat different remediation techniques. Spills to surface water are easier to clean up than spills to ground water, for obvious reasons. It is not only much harder to see the extent of the contamination, but also to remove the source of the contamination as, for example, a leaking underground storage tank. Since the underlying idea is to accelerate the rates of normal hydrocarbon biodegradation by overcoming the rate-limiting factors, the following concepts/techniques can lead to the results striven for:

- (i) Seeding with Microbial Cultures:* This is the approach often considered for the bioremediation of polluted environment, including soil and water bodies in a process termed bio-augmentation (Onyia, *et al.*, 2000). Most microorganisms considered for seeding are obtained by enrichment cultures from previously contaminated sites. However, because hydrocarbon-degrading bacteria and fungi are widely distributed in marine and freshwater habitats, adding seed cultures in some instances, has proven less promising for treating oil spills than adding

fertilizers and ensuring adequate aeration. Most tests have indicated that seed cultures are likely to be of little benefit over the naturally occurring micro-organisms at a contaminated site for biodegradation of the bulk of petroleum contaminants (Atlas, 1995).

(ii) Environmental Modification: Hydrocarbon degradation in marine environments is often limited by abiotic environmental factors such as molecular/dissolved oxygen, phosphate and nitrogen (ammonium, nitrate and organic nitrogen) concentrations. In well-aerated (high energy) marine environments, oxygen is, however, not limiting. Usually, marine waters have very low concentrations of nitrogen, phosphorus and various mineral nutrients that are needed for the incorporation into cellular biomass, and the availability of these within the area of hydrocarbon degradation is critical. This can be accomplished by adding nitrogen-containing fertilizers to the contaminated areas. In warmer climates where these nutrients could be washed away (and possibly even cause plankton blooms), slow-release fertilizers that are carrier-bound is adequate. Biodegradation of floating oil is limited by surface area. When a slick forms on the surface of the water, it may eventually disperse in the water column, aided by wave action or carried away for great distances. An option for stimulating biodegradation is to add a dispersant preferably containing a biosurfactant to the oil slick. This dramatically increases the surface area available for microbial colonization of the oil-in-water emulsion at the oil-water interface. The surfactants used in some dispersants have been shown to further enhance biodegradation of dispersed floating oil by serving as a biodegradable substrate and stimulating growth of biodegradative bacteria.

(d) Bioremediation of Groundwater Areas

Technologies used to remediate soils typically have equivalent versions for groundwater treatment. *In situ* techniques for treating groundwater typically involve injecting air and/or

nutrients to encourage indigenous bacteria to degrade the contaminants.

- (i) ***Pump-and-Treat:*** Pump-and-treat approaches involve bringing contaminated water to the surface and having it flow through a container or tank where treatment processes (bioremediation) can occur. Depending on the quality of the treated water, they can be discharged into the sewer system, surface water, or be reinjected into the ground. This technique works best in permeable soils. Non-homogeneous conditions in the subsurface may prevent clean-up of contaminant found in pockets.
- (ii) ***Gaseous Nutrient Injection:*** In this case, nutrients are fed to contaminated groundwater and soil via wells to encourage and feed naturally occurring micro-organisms. Vapour extraction is often used in conjunction with gaseous nutrient injection. The most common added gas is air. In the presence of sufficient oxygen, micro-organisms convert many organic contaminants to carbon dioxide, water and microbial cell mass. In the absence of oxygen, organic contaminants are metabolized to methane, limited amounts of carbon dioxide and trace amounts of hydrogen gas. Another gas that is added is methane. It enhances degradation by co-metabolism. That is, as bacteria consume methane, they produce enzymes that react with the organic contaminant and degrade it to harmless minerals.
- (iii) ***Oxygen Enhancement with Hydrogen Peroxide:*** An alternative to pumping oxygen gas into groundwater involves injecting a dilute solution of hydrogen peroxide (H_2O_2), which easily releases its extra oxygen atom to form water and free oxygen. This circulates through the contaminated groundwater zone to enhance the rate of aerobic biodegradation of organic contaminants by naturally occurring microbes. A solid peroxide product (e.g. oxygen-releasing compound, ORC) can also be used to increase the rate of biodegradation.

(iv) **Nitrate Enhancement:** A solution of nitrate is sometimes added to groundwater to enhance anaerobic biodegradation. On the other hand, high concentration of nitrate in surface waters and lakes can lead to phenomena known as algal bloom and eutrophication with time (2002).

(e) *Microbial Products of Importance in the Oil Industry*

A large variety of microbial surfactants (biosurfactants or bioemulsifiers) are produced by bacteria, yeasts and fungi. Biosurfactants are of increasing industrial interest because of their broad range of potential application, including emulsification, wetting, phase separation and viscosity reduction (Ochsner *et al.*, 1995). Almost all surfactants currently in use are chemically derived from petroleum. Interest, therefore, in microbial surfactants has been steadily increasing in recent years due to their diverse desirable characteristics such as selectivity effectiveness, environmental-friendly nature, stability at elevated temperatures, pH and salt concentrations. In addition to the above, biosurfactants have increased versatility in comparison to many synthetic surfactants and are suitable for production through fermentation. Their range of possible application in the oil industry processes alone includes cleaning up oil spills (both land and marine spills), removal of oily sludge from oil storage tanks through increased mobility as well as general enhancement of oil recovery processes from reservoirs and the level of the production well (Rosenberg, 1986).

The biopolymer, xanthan gum, has potential use in the petroleum drilling industry. High-performance circulating fluids formulated with bio-gums like xanthan gum reduce operating costs by increasing the penetration rates, reducing the torque and drag in high angle wells, improving the well bore stability, reducing friction pressure losses, fluid disposal costs, and minimizing the formation damage.

(f) *Plant Resources*

The clean-up of oil spills in certain environments, for example, coastal marshes, remains a problematic issue because wetlands can be extremely sensitive to the disturbances associated with

remediation activities. Foot traffic and equipment on the marsh surface during clean-up operations are considered secondary impacts of an oil spill that, in themselves, can cause significant harm by trampling vegetation, accelerating erosion, and burying oil in anaerobic sediments where it may persist for years (Getter *et al.*, 1984). It often appears that the best course of action may even be to let the marsh recover on its own to avoid these secondary impacts (Baker *et al.*, 1993; Mendelssohn *et al.*, 1993). Since it is known that wetland environments are sensitive to these and other impacts of clean-up, which can do even more damage than the spill itself, there is a need to develop alternative, less-intrusive oil spill clean-up bioremediation techniques that are compatible with the delicate wetland environment. These alternatives may be utilized without other clean-up methods in the event of a minor spill, or utilized in conjunction with existing clean-up methodologies in heavily oiled marshes.

Plant-based remediation (phytoremediation) is the use of plants to treat or stabilize contaminated soils, sediments or even groundwater. Some plant species have Parenchymatous tissue which facilitates the diffusion of oxygen from shoots to roots, and this ability varies between species (Drew, 1992). Therefore, different plant species provide different rhizosphere microenvironments, which, in turn, may affect the associated soil microbial community and the bioavailability of the oil to oxidative degradation. Plants can support the process of remediation in many ways:

- (i) **Rhizofiltration:** This is the removal of contaminants by adsorption to plant roots.
- (ii) **Rhizosphere Bioremediation:** This is the process of enhanced microbial degradation of contaminants near plant root surfaces.
- (iii) **Phytostabilization:** This involves erosion control of contaminated soils or hydraulic control through management of the soil water balance.

(iv) Phytoextraction: This refers to the removal and concentration of contaminants, generally metals, in plant tissues.

(v) Phytotransformation: This describes the process of plant uptake and transformation of contaminants to less toxic forms.

To be effective, phytoremediation projects must successfully integrate both the plant-based and engineering aspects of a system; this requires an interdisciplinary approach. A variety of grasses, alfalfa, clover and trees have reportedly been successfully used for phytoremediation (Childers *et al.*, 1997).

4.2.2.1. *Limitations and Concerns of Biological Processes in the Oil Industry*

- ◆ Under anaerobic conditions, contaminants may be degraded to a product that is more hazardous than the original contaminant.
- ◆ Introducing cold water or gas may slow the remediation process, as lower temperatures do not support degradation.
- ◆ Concentrations of hydrogen peroxide greater than 100 to 200 ppm in groundwater inhibit the activity of microorganisms. Safety precautions are also needed when handling hydrogen peroxide.
- ◆ Amended oxygen can be consumed very rapidly near the injection well, which creates two significant problems: biological growth can be limited to the region near the injection well, limiting adequate contaminant/microorganism contact throughout the contaminated zone; and biofouling of wells can retard the input of nutrients.
- ◆ Bioremediation is not well-suited for soils with low permeability (eg fine clays). High permeability is required to allow the nutrients reach the indigenous microorganisms.
- ◆ It is possible that the subsurface injection of gases below the water table can induce groundwater flow. It may be

necessary to use a pump-and-treat system in conjunction with gas injection for hydraulic control.

- ◆ The circulation of water-based solutions through the soil may increase contaminant mobility and necessitate treatment of underlying groundwater. If the process is enhancing groundwater bioremediation, a groundwater circulation system must be created so that contaminants do not escape from zones of active biodegradation.
- ◆ Nitrate injection to groundwater is of concern because nitrate is a regulated compound.
- ◆ Bioaugmentation using non-native micro-organisms is also controversial. It is feared that bioaugmentation with non-native microbes that are pathogenic will result in the multiplication and dissemination of the pathogen. This fear has been allayed by the use of edible yeasts from comestible natural sources such as palmwine (Amanchukwu *et al.*, 1988, 1989) as candidate micro-organisms in bioremediation.
- ◆ Very high contaminant concentrations may be toxic to both micro-organisms and plants.
- ◆ Micro-organisms can be a nuisance in stored petroleum products by causing chemical assimilatory biodeterioration and biofouling of such products (Okpokwasili and James, 1995).

4.2.3 Repositories and Microbial-Type Culture Collection

Infrastructural facilities and repositories are vital in different universities and research institutes to facilitate advanced research. Establishment of National Microbial-Type Culture Collection Centre is in progress in Nigeria. In the mean time, Universities and Research Institutes are encouraged to set up a network of manageable facilities for culture collection in their various establishments. In India, the repository on *Drosophila* has been strengthened to develop educational kits for demonstrating genetic experiments in schools and colleges.

4.2.4 A National Containment Facility

Establishment of a national containment facility is paramount for confined field trials of transgenic crops developed within Nigeria or imported from outside the country. Currently, the only available facility exists at the International Institute for Tropical Agriculture (IITA), Ibadan. USAID is considering the establishment of one at Sheda Science and Technology Complex (SHETSCO) for the trial of Bt-Cowpea being developed through the activities of AATF. This central facility will pave the way for the development and deployment of Transgenics to the benefit of the various stakeholders.

4.3 Agriculture/Food Security and Biotechnology

For the 4.6 billion people in developing countries, one billion do not get enough to eat and live in poverty. Twelve per cent (12%) of the world's land is arable and used for agricultural crops. It is however projected that the per capita availability of the land under agricultural use may be reduced from 2.06 hectares to 0.15 hectare by 2050.

Nigeria has a total landmass of 98.3 million hectares and a population of over 140 million. Over 70% of the population is basically agrarian (subsistence farmers). The cultivable land is 72 million hectares while 34 million hectares (47%) are actually cultivated. Thus, Nigerian agriculture is usually viewed as a land surplus economy. However, domestic food production has continued to lag behind the food needs of the population, making the country food-insecure. An alternative strategy needs to be sought for poverty reduction through food security. The industrialized countries of the West have scientifically established that an environmentally benign way of ensuring food security is through bioengineering of crops. What strategy or alternative is available for the developing countries?

Biotechnology has a lot to offer in the area of agriculture. It has been established as a new set of tools that will provide the breeders the capacity to improve agricultural productivity. Exploiting the development of new hybrids, including genes for abiotic and biotic resistance, is important. Sub-Saharan

African countries, of which Nigeria is one, did not experience Green Revolution and cannot therefore afford to be left behind in this era of Gene Revolution, which aims at improving crop productivity and quality through biotechnology. Developing planting material with desirable traits and genetic enhancement of all important crops will dominate the research agenda in the present millennium.

Nigeria is very rich in species and biodiversity. The country's bioresources should constitute the mainstay of the economy of the poor people if properly harnessed. Emphasis should be laid on plant biotechnology research, not neglecting animals. Molecular finger-printing, areas of genomics and proteomics have penetrated the barriers of fertilization to allow transfer of important traits from one plant to another. With the advent of gene transfer technology and its use in crops, plant biotechnologists have achieved higher productivity and better quality, including improved nutrition and storage properties. Adaptation of plants to specific environmental conditions such as increased plant tolerance to stress, pest and disease resistance, has been achieved for improved revenue generation.

Food security does not only involve availability of adequate quantity of food through improved yield; it also involves affordability of quality food, with high nutritive value to the citizens. Biotechnology therefore offers major inputs for healthier and more nutritious food. Most of the staple foods in the region are carbohydrate-rich but protein- and vitamins-poor; millions of people, particularly children, are malnourished. Vitamin A deficiency affects 40 million children worldwide and majority are in the developing countries. There are also serious deficiencies of iodine, iron, and other nutrients. A recent UNICEF report on food and nutrition deficiencies in children describes this as a "silent, invisible emergency with no outward sign of a problem." Every year, over 6 million children under the age of 5 die worldwide. More than half of these deaths result from inadequate nutrition. Promising leads are available in the areas of food fortification with required vitamins and other micronutrients to fight these nutritional deficiencies through application of biotechnology. A number of genetically improved

crops, otherwise known as transgenic plants, have been developed, tried and commercialized worldwide. Biotechnology application for the development of local crops is on-going in various laboratories across the country, and include, development of tissue culture regeneration protocols for important select species such as banana, plantain, cassava, pineapple, citrus, and neem, etc., which are expected to lead to major commercial activities; micropropagation technology, a tool also being employed for provision of high-quality planting materials to farmers. Other efforts include: development of *transgenic cassava*, African Biofortified Sorghum (ABS) and Bt-Cowpea which are currently undergoing confined field trials in various research institutes in the country; and *molecular mapping of cotton* to characterize its variability. Genetically improved foods, however, will have to be developed under adequate regulatory processes, with full public understanding. Safety and proper labelling of the genetically improved foods should be ensured so consumers will have a choice. This has been discussed under "biosafety issues."

Other areas of interest from the viewpoint of sustainable agriculture, soil fertility, and a clean environment and economic development include:

- (i) ***Establishment of a national facility*** for virus diagnosis and quality control in tissue culture raised planting materials with satellite centres in six geopolitical zones of the country. This facility will certify all domestic and export consignments.
- (ii) ***Biological Pest Control:*** Technologies for the mass production of candidate biocontrol agents, baculovirus, parasites, predators, antagonists, fungi and bacteria for economically important crops are available. The important crops include cotton, oilseeds, pulse, spices, sugarcane, tea, coffee and vegetables. Biopesticide from neem seeds have been developed by the National Research Institute for Chemical Technology (NARICT), Zaria. This technology has potential for commercialization.

- (iii) Insect Cell Culture:** Use of cell culture technique is an improvement on the biological control methods. Although the environmental advantages of biological control have long been recognized, manufacturing biological control products in marketable amounts has remained impossible. Insect cell culture removes these manufacturing constraints. In addition, like plant cell culture, insect cell culture is being investigated as a production method of therapeutic proteins (Layokun *et al*, 1988; Odeyemi *et al*, 1982 and Oloke, 1985).
- (iv) Biofertilisers:** Mass-production technologies for *Azotobacter*, *Azospirillum*, Blue Green Algae (BGA), *Mycorrhiza* and *Rhizobium* have been developed and are currently being marketed. *Rhizobium* biofertiliser has potential to save up to 25-30 kg chemical nitrogen in pulses and leguminous oil seeds. *Azotobacter*, *Azospirillum* and *Mycorrhiza* can make phosphate available in soluble form with other micronutrients to most of the commercial crops.

4.3.1 Plant Variety Protection

The protection of new plant varieties, especially agricultural crop varieties, is important to breeders, seed companies and users/growers of seed. Protection of new plant varieties is not as widespread as is the protection for innovations that can be copyrighted, trademarked or patented. A number of developing countries lack plant variety protection laws; however, some of these countries are now in the process of developing such laws. Much of this effort is under the guidance of the International Union for the Protection of New Varieties of Plants.

The International Union for the Protection of New Varieties of Plants (UPOV), an intergovernmental organization, was established to ensure that member states acknowledge the achievements of breeders by making available an exclusive property right, plant variety protection, based on a set of uniform and clearly defined principles. UPOV does not provide plant variety protection or certification itself, but assists countries in establishing plant variety protection principles and promotes international harmonization and co-operation

between member states. At present, there are 50 member countries. Sexually reproduced plants (plants grown from seed) are eligible for plant variety protection as well as tuber-propagated plant varieties. F1 hybrids can also be protected. Bacteria and fungi cannot be protected under this programme. In order to be considered for protection, a variety must be (i) *new*, available for less than 4 years in most counties, (ii) *distinct*, from all other varieties of the same "kind" of plant, (iii) *uniform*, in that all plants of the variety look the same, and (iv) are *stable*, in that they truly reproduce themselves – the plants are the same from generation to generation.

4.3.2 Animal Biotechnology

Livestock, like plant agriculture, is a key sector that should not be neglected. Livestock is a renewable resource that requires a sensitive treatment and a careful management to enable it reach its full productive potential. The first Nigerian National Livestock Resource Census was conducted in 1990 under the auspices of the Federal Livestock Department (FLD) in collaboration with Resource Inventory and Management Limited (RIM) as a special study within the Second Livestock Development Project (SLDP), which was jointly financed by the World Bank and the Federal Military Government of Nigeria. The census, which was carried out using low-level aerial surveys and Systematic Reconnaissance Flight (SRF) technique, provided the first review of the species, varieties and production systems of domestic livestock in Nigeria. The report of the survey reveals that the major livestock of the country and their population estimates are as follows: poultry, 102,832,000 (excluding birds in commercial poultry farms, but including 15 million pigeons); goats, 34,495,000; sheep, 22,104,000; cattle, 13,947,000; pigs, 3,410,000; donkeys, 936,000; horses, 208,000; camels, 88,000 (see Table 4.3 for more details). The above record is not too different from the recent (2007) report given by the Department of Livestock and Pest Control Services, which put the current poultry population at 140 million distributed sectorally as follows: Commercial poultry, 25%; Semi-commercial poultry, 15%; and Backyard poultry rearing, 60%.

A similar report on the poultry status in China reveals that population of poultry is about three times that of humans in China (i.e. 3:1), whereas Nigeria's stands at 1:1 going by the current census figure conducted in 2006. The low populations of other bigger species of livestock are even more discouraging, as some of the species like camels and horses are close to extinction. This situation can explain why animal protein in form of meat and milk, is largely unaffordable to the majority of the poor masses of Nigeria.

Table 4.3: Compendium of Nigerian Livestock Population Estimates

Year	Cattle	Goats	Sheep	Pigs	Donkeys	Horses	Camels	Poultry
(in thousands)								
Northern Nigeria Veterinary Department: (various years)								
1950	3,732							
1952	3,689							
1953	4,070							
1954	4,155							
1955	4,071							
1956	3,867							
1957	3,837							
1958	4,143	4,871	2,327	72	750	231	3	
1959	3,992	6,667	2,443	68	624	245	5	
1960	4,173	5,040	2,637	96	954	246	3	
1961	4,467	5,679	5,831	96	823	309	5	
1962	4,256	6,359	3,482	91	766	279	4	
Federal Office of Statistics: (FOS, various years)								
1960	4,428	14,990	5,080	596	1,500	310		32,920
1977	3,449	21,919	6,391		1,036			66,330
1982	7,494	28,851	13,687	672	1,200			71,060
1983	6,679	36,745	15,664	658	1,371			76,085
1984	5,233	27,706	13,915	491	968			69,434
1985	5,110	26,148	12,193	643	889			68,854
1986	6,755	24,578	13,049	864				74,707

United Nations Food and Agriculture Organization: (FAO, various years)								
1963	10,859	21,206	7,235	680	2,085	431	17	66,040
1975	11,000	22,500	7,650	880	720	250	18	85,000
1979	12,000	24,000	11,350	1,100	700	250	17	110,000
1980	12,300	24,000	11,700	1,100	700	250	17	120,000
1985	12,000	26,000	12,850	1,300	700	250	18	160,000
1989	12,200	26,000	13,200	1,300	700	250	18	200,000
National Livestock Survey: (This study)								
1990	13,947	34,495	22,104	3,410	936	208	88	102,832*

* Excludes birds in commercial poultry farms, but includes 15 million pigeons

4.3.2.1 Importance of Livestock to the Nigerian Economy

Cattle, sheep, goats and pigs are the primary source of animal protein for the whole nation. Beef is the favourite meat all over the country but the Moslems also enjoy mutton. Goat meat, on the other hand, is a favourite of many people in the Middle Belt and South-eastern states, while in the same regions only a few people appreciate pork as a delicacy (Ariyo, 1998). The urban population of the northern pastoral zones depend on cattle as the primary source of milk and milk products. Hides and skins from cattle, sheep and goats were an important export from northern Nigeria to Europe, bringing in over five million pounds sterling in the pre-civil war years (Ariyo, 1998). Since the post-Nigerian civil war, there has been hardly any export of hides and skin because of the large domestic demand for them.

4.3.2.2 Factors Affecting Livestock Development

A number of factors plague the development of livestock in Nigeria. These include the neglect of Research and Development (R & D) and the deployment of research findings to this sector. Most of the causes of death to animals are preventable or curable. According to the above report, causes of death include: lung infections, rinderpest, dysentery, diarrhoea, oedema/broken leg, anaemia caused either by trypanosome or *Babesia bygemina*, lack of milk, abortion/

stillbirths, and absence of bone marrow. More devastating are the issues of epidemic outbreaks like those of swine fever, avian flu, foot and mouth disease, mad cow disease, etc. Unchecked introduction of livestock from across the borders can also bring about introduction of disease which the country does not have immediate solution for.

A number of research institutes have been established with specific mandates for animal production and disease control. These research institutes include: National Animal Production Institute and National Veterinary Research Institute. Their major constraint has always been inadequate funding. A number of livestock species in Nigeria have been gradually wiped out. This trend, if not scientifically (best still, biotechnologically) checked, will lead to serious loss of bioresources and biodiversity.

4.3.3 Fishery

Nigeria is blessed with extensive inland water mass estimated at about 12.5 million hectares (waters of Rivers Niger, Benue, Gongola, Argungu, Komadugu, Yobe and of Lakes Chad and Kainji, to mention but a few), capable of producing over 5,000,000 metric tons of fish annually under proper management. Small-scale fishermen using traditional fishing gear and watercraft dominate the fishing industry. These fishermen are responsible for over 80 per cent of the annual fish output of about 500,000 metric tons from rivers and lakes, creeks and lagoons and the Atlantic coastal waters. This amount of fish represents less than a quarter of the nation's annual requirement. The fishes are delivered fresh daily to nearby markets while distant markets are supplied with low-quality smoked or sun-dried fishes. Domestic fish output is supplemented by imports of frozen fish from Europe, South America and the Far East. The government is taking active interest in the fishing industry by directing efforts towards increasing the offshore catch and extending the inland areas where fish can be caught. The government is also trying to improve the fish stocks of the Nigerian waters, modernize the country's fishing industry, and empower the Nigerian fishermen to take to the sea in order to increase the

domestic output of fish and save precious foreign exchange now going for importing frozen fishes of doubtful quality. The National Biotechnology Development Agency (NABDA) has further improved the fish stock by introducing re-circulatory fish farming and fingerling rearing (which had hitherto been imported). Another action to be taken seriously by the government is that of commercial production of quality floating feed within the country. Viable fish farming has been hindered by low-quality feed that are largely unavailable to the fish because they sink fast before the fish can catch them.

4.3.3.1 Scientific / Biotechnology Intervention

(i) Mammalian Cell Culture: Mammalian Cell Culture, as an essential tool for livestock breeding, has been in use for decades. The process involves eggs and sperm, taken from genetically superior animals (bulls and cows), being united in the laboratory and the resulting embryos grown in culture before being implanted in a surrogate mother. A similar form of mammalian cell culture has also been an essential component of the human *in vitro* fertilization process.

India is one developing country that has revolutionized her livestock and dairy industry through the application of biotechnology. Eighty per cent (80%) of the milk in India come from small and marginal animal farms, leading to a major social impact. India established a diverse infrastructure to help farmers in the application of embryo transfer technology. The world's first IVF buffalo calf was born through the embryo transfer technology at the National Dairy Research Institute. Multiple ovulation and embryo transfer, *in vitro* embryo production, embryo sexing, vaccines and diagnostic kits for animal health, have also been developed.

Nigeria can borrow a leaf from both India and China to improve on the current livestock status through the application of biotechnology. The long stretch of coastline along the Atlantic Ocean, offers the country a great potential for marine resource and aquaculture development. To improve her annual production of fish, scientific aquaculture in form of re-circulatory fish farming offers great possibilities. In fact, aquaculture products

are among the fastest-moving commodities in the world. To achieve substantial and sustainable increased diversified nutritional intake of the citizenry, scientists have to continuously improve seed production, feed, health products, cryopreservation, genetic studies, and related environmental factors. This is a sure way to combat nutritional deficiency.

(ii) Establishment of Bioresources Development Centres (BIODEC): Establishment of Bioresources Development Centres in the six geopolitical zones corresponding to the six ecological zones in the country is in the forefront of NABDA's programmes. One of the objectives of this programme is for extension of technologies which are pro-poor, pro-women and pro-nature to the grass roots level as a model for rural development. Farmers of target communities are trained to acquire skills in their areas of natural trade or interest. One of such centres that has been fully established is located at Odi, Bayelsa State in the South-South zone of the country. The main occupations of the people are farming and fishing, which have been disrupted by the activities of oil companies due to incessant oil spills that have polluted both land and water causing serious environmental degradation. Some of the skills being taught to the local people in the area include: modern fish farming (including fingerlings production), using ponding or concrete tank systems; grass-cutter rearing; snailry; and mushroom production with application of tissue culture techniques. These skill acquisition training programmes are designed to encourage women and youth entrepreneurs to take up bio-enterprises that empower them for wealth creation and poverty alleviation.

Other programmes of NABDA targeted towards achieving the Millennium Development Goals aimed at environmental sustainability and improved sanitation at the grass roots level include: production and use of biogas as alternative to woodfuel as energy source for cooking; bio-fertilizers and bio-pesticides (as against chemical-based products that pollute the environment); Ecological Sanitation (EcoSan), which provides

farmers with the option and opportunity for safe use of sanitized human waste for improved agricultural yields.

4.4 Medical Biotechnology, Pharmaceuticals and Industry

A major responsibility of biotechnologists in the 21st century is to develop low-cost, affordable, efficient and easily accessed health care delivery systems. Sustainable exploitation of the country's bioresources is one of the several options. Advances in biotechnology can help in converting bioresources to useful products, processes and technologies through the adoption of multi-disciplinary approach. The pharmaceutical sector has had a major impact in this field, as rare therapeutic molecules in the pure form become available. Diagnostics have expanded, with over 600 biotechnology-based diagnostics (valued at about US\$20 billion worldwide) now available in clinical practice. The polymerase chain reaction (PCR)-based diagnostics are the most common. According to records from Andes Pharmaceuticals (USA), a company specialized in bioprospecting for anti-cancer drugs, 40 per cent of all prescription drugs in the USA are based on natural products. In the '90s, half of the 20 most sold medicinal products in the world were developed from natural product research, with total annual sales of around US\$ 15 billion. By 2002, the market for plant-derived drugs had grown to US\$ 30 billion, which would rise to more than 60 per cent, if anti-cancer drugs are considered. Bioprospecting for drug development in developing countries has therefore become very attractive to pharmaceutical companies, as developing countries are not only rich in biodiversity, but also in traditional knowledge and healing practices. Globally, there are about 35-40 biotechnology-derived therapeutics and vaccines in use and more than 500 drugs and vaccines in different stages of clinical trial. The global market for herbal products has been approximated to reach around US\$5 trillion by 2050 (Sharma, 2004). The production of pharmaceuticals is *research and development* (R&D)-based and it is estimated that R&D budgets of leading companies account for 10 to 15 per cent of their annual sales. If parts of these expenses were spent on biodiversity prospecting in developing countries, equitable sharing of

benefits as it is foreseen by the CBD could help these countries to create new sources of revenue and to extend their scientific capabilities.

Nigeria is rich in bioresources but bioprospecting for drug discovery is still in its infancy. The Nigerian Natural Medicine Development Agency (NNMDA) has carried out surveys on medicinal plants in some parts of the country, but there is need for development of a national programme involving other stakeholders and the private sector (particularly the pharmaceutical companies) for Nigeria to start reaping the benefits in this sector.

4.5 Bioinformatics and Biodiversity Conservation and Data Management

Biodiversity encompasses the taxonomic and functional diversity of living organisms. In biodiversity, we need documentation, digitalization, networking, integration and co-ordination to achieve individual and organizational goals. An advanced, integrated, planned, organized data management, information process and dissemination system is useful in implementing environmentally sound and sustainable utilization of the nation's biological resources. Such information system can help in making decisions, policies and plans, and in implementing strategies.

Bioinformatics is the application of computers and information technology (IT) for collection, collation, storage, analysis, modelling, simulation and dissemination of diverse and distributed data/information about biotic resources and their environment. Hence, bioinformatics is described as the tools and techniques for storing, handling and communicating the massive and ever-increasing amounts of biological data emerging principally from genomics research.

Before the emergence of bioinformatics, data storage and retrieval were the nightmare of systematics researchers; but bioinformatics now enables biodiversity researchers to communicate efficiently with one another, and provide a platform and a common language which helps to put biodiversity information into the public domain in accessible forms on the

internet. The truth of the matter is that "bioresources and diversity are available in developing countries but the information lies with developed countries."

The application of bioinformatics in biodiversity is important for the following reasons:

- (i) The complex and voluminous data of biodiversity can be digitalized for easy accession, analysis and interpretation.
- (ii) It facilitates the survey, documentation and measurement of biodiversity data.
- (iii) Based on the available data, future biodiversity of a particular area can be predicted and a model formulated by computational methods; thus, appropriate measures can be taken for its conservation and sustainable utilization.
- (iv) It helps to predict species invasions using ecological niche modelling.
- (v) The electronic information may serve as the raw material for augmenting future developments in all areas of biology.
- (vi) The digital databases can easily provide the current status of the biodiversity of a particular area.
- (vii) The biodiversity extinction rate can be easily documented while theoretical studies and modelling can be formulated for its conservation on a priority basis.
- (viii) The computation analysis makes for easy understanding of the phylogenetic relationship among the individual species.
- (ix) Through the internet, biodiversity databases can be linked together and the information shared.
- (x) Researchers can easily identify the priority materials for their studies.
- (xi) The potential indigenous material can be easily identified for biotechnological intervention.

The coming together of biotechnology and informatics is paying rich dividends even in other sectors apart from biodiversity identification and management. The number of genes characterized from a variety of organisms and the number of evolved protein structures are doubling bi-annually. Genome

projects, drug design, and molecular taxonomy are all becoming increasingly dependent on information technology while information on nucleotides and protein sequences is accumulating rapidly.

Bioinformatics development is one of the key mandates of the Nigerian National Biotechnology Development Agency, which has established a technical Department of Molecular Biology and Bioinformatics to oversee all projects relating to molecular biology and bioinformatics, and to ensure the evolution of a national bioinformatics network for internet-based management of the nation's huge biological data resources among others. It is expected to establish link/access to other internationally recognized databases such as Genebank, EMBnet, PDB, GDB, EBI and Plant Genome database. These databases are being utilized for genomic and proteomic R&D activities through high-speed and high-bandwidth network services.

4.6 Intellectual Property Rights

The overall objective of Intellectual Property Rights (IPR) is:

To promote the progress of arts, sciences and technology, by securing for limited times to authors and inventors the exclusive rights to their respective writings, knowledge and discoveries.

Intellectual Property is intangible personal property that may be legally protected if appropriate steps are taken. The categories of Intellectual Property (IP) are Patents, which protect new, useful, and "non-obvious" inventions and processes; Copyrights, which protect original works of authorship; Trade Marks, which protect words, names, and symbols used to identify goods and services; and Trade Secrets.

Currently, the issue of intellectual property has moved to the international arena. According to the World Trade Organization (WTO)'s rules on intellectual property, it is obligatory for all member countries to implement minimum standards of IP for plant varieties and micro-organisms. Nigeria is a member of the World Trade Organization.

(i) Nigerian IP Laws

- ◆ Patents and Designs Decree No. 60 of 1970;
- ◆ National Office of Industrial Decree No. 70 of 1979;
- ◆ Trade Marks Act No. 29 of 1965;
- ◆ Copyright Decree No. 47 of 1988, No. 98 of 1992, and No. 42 of 1999.

(ii) Nigeria's IP membership

Nigeria is either a member or signatory to the following pacts:

- ◆ Paris Convention since 1963;
- ◆ Berne Convention since 1993;
- ◆ Rome Convention since 1993;
- ◆ Member of WIPO since 1995;
- ◆ Patent Law Treaty since 2000.

(iii) Government Ministries/Agencies Regulating IPR in Nigeria

- ◆ Federal Ministry of Commerce, Commercial Law Department;
- ◆ Federal Ministry of Commerce, Registry of Trade Marks, Patents and Designs;
- ◆ National Office for Technology Acquisition and Promotion (NOTAP).

4.6.1 Plant Protection and Patents

(a) Patent

Inventions are protected through patents. An invention is an innovation that has three characteristics, namely: (i) novelty, meaning it is new to the patent literature; (ii) non-obviousness to one skilled in the science or practical applications of the innovation; and (iii) utility or usefulness. Inventions cover many areas, including chemistry, medicine, machinery, vehicle parts, computer technology, computer programmes, and biotechnology.

Patents rights are obtained from a governmental patent office where application materials and forms are also available. Often, a patent attorney is used to draft the patent application and complete the appropriate forms. Patent attorneys are skilled in reviewing the patent literature that is necessary to

prove the novelty of the invention. Attorneys are able to use the information provided by the innovator, and this is learnt through literature search to draft an application that provides the broadest protection for an invention. One important part of a patent is the section termed "Claims." The "Claims" are what the invention can do – the patent claims it can do these things. The claimed areas are what the patent protects.

(b) Plant Variety Protection (PVP)

The International Union for Protection of New Varieties of Plants (UPOV)

- ♦ UPOV was ratified in 1968 and followed with conventions in 1972, 1978 and 1991.
- ♦ *Characteristics of a new variety: the DUS standards*
The new variety should be:
 - Distinct,
 - Uniform, and
 - Stable.
- ♦ *Conditions for Plant Variety Protection*
 - Basic right is to be able to exclusively market the variety for a limited period of time:
 - 20 years for herbaceous plants,
 - 25 years for trees and vines.
 - Farmers' Rights to be observed.
 - Application to be prepared and filed.
 - Sample of seeds to be included.
 - Protecting agency may use seeds to determine if DUS is met.
 - Protection can be granted individually in each country.
 - Ability to restrict seed sales through licences.
 - Ability to restrict breeding due to infringement.

U.S. is the only country that allows plant patents but almost every country has some sort of Plant Breeder's Rights (PBR).

(c) Farmers' Rights

- ♦ Parties should recognize the contribution of farmers.

- ♦ The responsibility for realization of right rests with national governments.
- ♦ Each party (in accordance with needs and priorities and as appropriate) shall take measures to protect and promote:
 - Traditional knowledge,
 - the right to participate in benefit sharing, and
 - the right to participate in decision-making by adopting a bottom-up approach.

The Article above does not limit *“any rights that farmers have to save, use, exchange and sell farm-saved seed / propagating material subject to national law and as appropriate.”*

(d) Traditional Knowledge

- ♦ What is included in this category?
 - Medicinal, agricultural and ecological knowledge;
 - Music, dance, poetry and stories;
 - Artistic endeavours;
 - Spiritual expression.
- ♦ No law is governing traditional knowledge currently
- ♦ Traditional knowledge is passed on orally across generations.
- ♦ There is collective ownership.

In Nigeria, a steering on IPR and TKS (i.e. Traditional Knowledge System) under the aegis of the Nigerian Natural Medicine Development Agency has currently been set up to prepare a draft bill on IPR and TKS for eventual passage into law.

(e) Ethical Issues

The bioethics committee of UNESCO established in 1993 has evolved guidelines for ethical issues associated with the use of modern biotechnology.

4.7 Biotrade Initiative

Biotrade Initiative is a collaborative effort that involves the Secretariat of the CBD and several United Nations agencies such as the United Nations Conference on Trade and Development (UNCTAD). The argument that biodiversity conservation can be promoted through its profitable use

engendered the setting up of an international framework to create new 'bio-partnerships' known as *Biotrade Initiative*. This programme offers opportunities for developing countries to sustainably use and commercialize bioresources. However, its use as an engine for economic growth in developing countries has been limited by institutional and macroeconomic constraints. This international treaty aims at the conservation of biological resources, the sustainable use of their components and the fair and equitable sharing of the benefits arising from their use. The launching of Biotrade Initiative by the third *Conference of the Parties* (COP) of the CBD in November 1996 was to enable developing countries that are rich in bioresources to use funds generated from bioresources for their country's development.

4.7.1 Promotion of Profitable Use of Bioresources under Biotrade Initiative

The Initiative has advocated the collaboration of all relevant stakeholders, including those perceived as potential rivals, both in industrialized and developing countries: the private sector, including both multinational corporations and local companies; governments; universities; financial institutions; and local and indigenous communities. If successfully implemented, co-operation could take place both at the local community and national levels.

The Core Activity Areas

The core activity areas of the initiative cover:

- ♦ ***Bioprospecting:*** Genetic resources are screened for commercially valuable chemicals and genetic compounds.
- ♦ ***Non-timber Forest Products (NTFPs):*** These are Biological resources other than timber that can be harvested from forests for own use and/or for trade. Examples of NTFPs include medicinal plants, fibres, resins, oils and gums.
- ♦ ***Eco-tourism:*** Richness in biological resources, including wildlife, can be attractive for the development of tourism. Eco-tourism is based on biological diversity and its use in a sustainable manner.

4.8 Biosafety Issues

Sustainable exploitation of bioresources with application of modern biotechnology tools has been associated with certain risks. Some of the risks are real, while others are imagined. These risks fall into two basic categories with the effects on human and animal health as well as the environmental consequences expressed below:

- ◆ The risks of transferring toxins from one life form to another, or creating new toxins or of transferring allergenic compounds from one species to another which may result in unexpected allergic reactions,
- ◆ The most publicized health risk is that GM crops carrying antibiotic genes used as markers may generate antibiotic resistance in livestock or humans, while the most serious environmental risk is the likelihood of transgenes escaping from cultivated crops into wild relatives. This may lead to the development of more aggressive weeds with increased resistance to diseases or environmental stresses, upsetting the ecosystem balance.
- ◆ Biodiversity may be lost, due to the displacement of traditional cultivars by a small number of genetically modified cultivars.
- ◆ There is a potential risk for pests to evolve resistance to toxins produced by microbe genes, e.g. the transfer of some genetic materials to corn and cotton from *Bacillus thuringensis (Bt)* which produces insecticidal toxin.
- ◆ Another potential hazard arises from plants containing genes from viral pathogens that confer resistance to these same pathogens.
- ◆ There are fears that have less scientific basis. For example, there is no convincing evidence to date that the process of gene transfer confers a health risk.
- ◆ There is also no reason why ingesting pieces of transgenic DNA is likely to be hazardous, any more than the large quantities of DNA from numerous sources ingested everyday in normal diet.

4.8.1 Alleviation of Fear

Modern Biotechnology therefore needs to be regulated for the following reasons:

- ◆ To achieve the following landmark objectives of the Convention on Biological Diversity (CBD):
 - The conservation of biological diversity;
 - The sustainable use of its components;
 - The fair and equitable sharing of its benefits arising out of utilization of genetic resources.

Objectives of Cartagena Protocol Biodiversity (CPB)

- ◆ In accordance with the precautionary approach contained in Principle 15 of the Rio Declaration on Environment and Development, the objective of the Cartagena Protocol Biodiversity is to contribute to ensuring an adequate level of protection in the field of the safe transfer, handling and use of living modified organisms resulting from modern biotechnology that may have adverse effects on the conservation and sustainable use of biological diversity, taking also into account risks to human health, and specifically focusing on transboundary movements in a regulatory process known as biosafety.

4.9 Biosafety

Biosafety is a term used to describe efforts and steps employed to reduce and eliminate the potential risks resulting from modern biotechnology and its products (UNEP, March 2002).

4.9.1 The Evolution of Biosafety in Nigeria

Modern biotechnology regulation in Nigeria started in the early 1990s as researchers in modern biotechnology became competent. The development of a biosafety regime has followed an evolutionary pattern. Each successive step is an improvement on the preceding level taking cognisance of our national and international obligations in the field of biosafety.

4.9.2 The Cartagena Protocol on Biosafety

The Convention on Biological Diversity (CBD), which Nigeria signed in 1992, identified the emergence of Genetically

Modified Organisms (GMOs)/Living Modified Organisms (LMOs) as a group of items produced by modern biotechnology that needed special attention because of their potential adverse impacts on biodiversity and human health.

The Convention therefore recommended that a protocol to control the trans-boundary movement of GMOs be negotiated as soon as possible. An instrument, the Cartagena Protocol on Biosafety (CPB) was eventually negotiated and became available for signature in 2000. It was signed by Nigeria in 2000 and ratified by the then President, Chief Olusegun Obasanjo on 30th November, 2002. The protocol came into force on 11th September, 2003.

The protocol's major plan is to contribute to ensuring an adequate level of protection in the field of safe transfer, handling and use of living modified organisms resulting from modern biotechnology which may have adverse effects on conservation and sustainable use of biodiversity, taking into account risks to human health and specifically focusing on trans-boundary movements.

4.9.3 Biosafety Guidelines

Nigeria has developed two Biosafety Guidelines since 1994.

(a) 1994 Biosafety Guidelines

These guidelines only covered agricultural modern biotechnology and were not actually approved by the government and these were before the advent of the Cartagena Protocol on Biosafety (CPB).

(b) 2001 Biosafety Guidelines

These guidelines were approved by the Federal Executive Council as a fast-track approach to encourage the development and regulation of modern biotechnology in the country having signed an internationally binding protocol. The guidelines are more encompassing in all biosafety issues beyond agricultural modern biotechnology in line with the Cartagena Protocol on Biosafety.

The Biosafety Guidelines are however not legally binding and contain some gaps. Consequent upon this, there was the need to develop a policy and law on Biosafety that would address

the subject of regulating modern biotechnology using a more robust and encompassing approach. The Federal Executive Council approved the development of a bill on biosafety and a national committee was inaugurated in 2002 for that purpose.

4.9.4 Development of National Biosafety Framework (NBF)

In recognition of the need for protection of human health and the environment (biodiversity) from potential risks arising from the practice of modern biotechnology and the handling and use of its products, the Federal Government inaugurated a National Co-ordinating Committee to draft the National Biosafety Framework under the UNEP/GEF Development Project on 2nd December, 2002.

4.9.5 National Co-ordinating Committee

The Committee, which commenced work immediately, formally submitted the following outputs of the development of the National Biosafety Framework (NBF) to the Honourable Minister of Environment, Housing and Urban Development on 21st November, 2006:

- ♦ National Biosafety Policy;
- ♦ Biosafety Draft Bill;
- ♦ Public Awareness and Participation in Biosafety;
- ♦ Request and Authorization.

The NBF is to ensure the safety of modern biotechnology practice, the handling and use of its products (genetically modified organisms) that may have adverse effects on conservation and sustainable use of biodiversity, taking into account risks to human health.

4.9.6 Scope of Framework

- ♦ The Nigerian National Biosafety Framework covers all modern biotechnology activities, genetically modified organisms (GMOs) and the products of modern biotechnology;
- ♦ Laboratory and field applications of modern biotechnology, currently known to science as well as those that may be developed in future;

- ♦ Current and future applications of modern biotechnology in agriculture, human and veterinary medicine, food/feed and beverage production, industry, environmental management, (bioremediation, industrial and domestic waste management, etc.)

4.9.7 Regulatory Regime

These include:

- ♦ Notification;
- ♦ Information transfer and review;
- ♦ Risk assessment, including socio-economic impact and ethical consideration;
- ♦ Monitoring and enforcement measures relevant to import, export, transboundary movement of the products of modern biotechnology, laboratory and field testing/use of modern biotechnology, including handling, containment, disposal, control, monitoring and release;
- ♦ Ensuring that any GMOs that might be eaten by humans and/or animals are safe to eat;
- ♦ Research and development in modern biotechnology, including applications in academic, agricultural, industrial and other categories of research;
- ♦ Occupational safety in the workplace where modern biotechnology procedures are used or products handled;
- ♦ Labelling of genetically modified organisms (GMOs) in food/feed produced locally, sold domestically or imported;
- ♦ Any other measures that may be required for safe use of modern biotechnology while protecting human health, the environment and national biodiversity;
- ♦ Promotion of public awareness on biosafety involving policy-makers, legislators, administrators, the organized private sector, industry and the rural community;
- ♦ Development and establishment of a comprehensive and up-to-date scientific database and infrastructure for information exchange to enable risk assessment and

evaluation of products and provision of a mechanism for effecting advanced informed agreement.

4.10 Framework Implementation Strategy and Institutions

The framework seeks to facilitate the establishment and development of national capacity to assess and manage potential risks associated with modern biotechnology practice and products. Establishment of well-coordinated and sustainable mechanisms and institutional structures for the effective implementation of the Biosafety Framework have and are being put in place in Nigeria. The structure comprises, but is not limited to:

- ♦ National Focal Point /Competent National Authority on Biosafety (Federal Ministry of Environment, Housing and Urban Development) already in place;
- ♦ National Biosafety Committee (NBC) already in place;
- ♦ Technical Sub-Committee(s) – NBTS to be constituted as the need arises;
- ♦ Institutional Biosafety Committee(s) – IBC, mandatory for institutions that apply modern biotechnology practice.

4.10.1 National Focal Point / Competent National Authority

The Federal Ministry of Environment, Housing and Urban Development, serving as the National Focal Point, is responsible for liaison with the Secretariat of the Convention on Biological Diversity (CBD), for the administrative functions required under the Cartagena Protocol on Biosafety. The Ministry, which also serves as the Competent National Authority for Biosafety in Nigeria, is responsible for the safe management of modern biotechnology activities, including research, development, introduction and the use of the products of modern biotechnology (GMOs).

4.10.2 National Biosafety Committee (NBC)

NBC was established to advise the Biosafety Competent National Authority on matters relating to the safe practice of

modern biotechnology and the handling, transfer and use of products of modern biotechnology.

4.10.3 Sub-committees

These would be established as the need arises by the NBC for sectoral interests such as agriculture, health, industry and environment.

4.11 Development of National Biosafety Clearing House

The UNEP/GEF has signed MOU with Nigeria to develop Biosafety Clearing House Capacity. The Biosafety Clearing House is to facilitate the exchange of scientific, technical, environmental and legal information, and experience with GMO/LMO (reference: <http://www.bch.biodiv.org>).

This project is an add-on to the recently completed National Biosafety Framework project.

The project's duration was twelve (12) months from March 2007 but subject to renewal. The focus of the project is provision of infrastructure and training for developing information on Biosafety.

4.11.1 Project Output

The expected outputs at the close of the project include:

- (i) Information Communication Technology (ICT) equipment;
- (ii) Functional National Biosafety Website that has linkages with the Convention on Biodiversity, Central Portal and portals of relevant National Institutes/Agencies.
- (iii) Improved awareness of the importance of the Cartagena Protocol on Biosafety and Biosafety Clearing House (BCH);
- (iv) A corps of trained Nigerians in strategic government ministries and agencies who can find information and be able to send information into the global BCH system;
- (v) Improved provision of information infrastructure that will enable Nigeria to participate in the BCH network and access to information by the public;

- (vi) Creation of relevant Biosafety Data Bank (roster of biosafety experts, national institutions with capacity for biotechnology).

4.12 Nigeria's Current Position with Biosafety Bill

- ♦ The Draft Biosafety Bill is currently being sponsored as a private member bill at the National Assembly. It has gone through the first and seconding readings and public hearing at the House of Representative and now ready to be presented for final reading at the senate.

4.12.1 Other Legislative Instruments

- ♦ Seed Policy;
- ♦ Phytosanitary Legislation; and
- ♦ Food Law and Legislation (Codex Alimentarius).

References

- Amanchukwu, S.C., Obafemi, A. and Okpokwasili, G.C. (1989). "Hydrocarbon Degradation and Utilization by a Palmwine Yeast Isolate." *FEMS Microbiol. Lett.* 57: 151 – 154.
- Amanchukwu, S.C., Okpokwasili, G.C. and Obafemi, A. (1988). "Factors Affecting Hydrocarbon Degradation by *Schizosaccharomyces Pomnbe* Isolated from Palmwine." *The Petroleum Industry and the Nigerian Environment, Proc 1987 International Seminar*, pp. 97 – 103, NNPC, Lagos.
- Atlas, R.M. (1995). Petroleum Biodegradation and Oil Spill Bioremediation. *Mar. Poll. Bull.* 31: 178 – 182.
- Baker, J.M., Guzman, L.M. Bartlett, P.D. P.D. Little, P.D. and Wilson, N.C. (1993). "Long-term Fate and Effects of Untreated Thick Oil Deposits on Salt Marshes. *Proceeding: of the 1993 International Oil Spill Conference*, American Petroleum Institute, Washington, D.C., pp. 395 – 399.
- Blackburn, J.W. and Hafker, W.R. (1993). "The Impact of Biochemistry, Bioavailability and Bioactivity on the Selection of Bioremediation Techniques." *Trends Biotechno.* 11: 328-333.
- Charrier, A., Fridlansky, F. and Mounolou, J. C. (1996). "Biological Diversity and Genetic Resources." In *Biodiversity, Science and Development: Towards a New Partnership*. (di Castri, F and Younnès T. (eds). Cab International, Oxon OX10 8DE, UK.

- Childers, G.W., Shafer, G. P. and Dowty, R.A. (1999). "Establishing a Protocol for Bioremediation of Small-scale Oil Spills in Louisiana." Louisiana Applied Oil Spill Research and Development Programme. OSRADP Technical Report Series 96 – 003.
- Choi, D.H., Hori, K. Tanji Y. and Unno, H. (1999). "Microbial Degradation of Solid Alkane in Non-degradable Oil Phase." *Biochemical Engineering Journal* 3: 71 – 78.
- Conklin, A.R. (1997). Scientists seek bioremediation answers. *Soil and Groundwater Clean-up*, pp. 27 – 29.
- Drew, M.C. (1992). "Soil Aeration and Plant Root Metabolism. *Soil Science*. 154: 259 – 268.
- Environment Canada (1995). Bioremediation of Petroleum-contaminated Soils: An Innovative Environmentally Friendly Technology. Evaluation and Interpretation Branch, Ottawa, Ontario.
- Foght, J.M. and Westlake, D.W.S. (1989). "Biodegradation of Hydrocarbons in Freshwater." In *Oil in Freshwater: Chemistry, Biology, Countermeasure Technology*. J.H. Vandermeulen and S.E. Hrudey. Pergamon Press, London , pp. 217 – 230.
- for the Acquisition of Agri-biotech Applications.
- Fowler, C. and Mooney, P. (1990). *Shattering Food, Politics, and the Loss of Genetic Diversity*. Tuscon: The University of Arizona Press, p. 278.
- French, C.E., Rosser, S.J. Davies, G.J. Nicklin, S. and Bruce, N.C. (1999). Biodegradation of Explosives Bytransgenic Plants Expressing Pentaerythritol
- Getter, C.D., Cintron, G. Dicks, B. Lewis III, R.R. and Seneca, E.D. (1984). "The Recovery and Restoration of Salt Marshes and Mangroves Following an Oil Spill. In: J. Cairns, Jr. and A L. Buikema, Jr. (eds.), *Restoration of Habitats Impacted by Oil Spills*, Butterworth Publishers, Boston, pp. 65 – 105.
- Global Food Security* (ed.) M.S.Swaminathan and S.Jena Macmillan Publication,
- Harrington, C., Dobell, D., Raphael, M., Aubry, K., Carey, A., Curtis, R., Lehmkuhl, J. and Miller, R. (1990). *Stand-level Information Needs Related to New Perspective*. Pacific Forest and Range Experiment Station, Olympia Forestry Sciences Laboratory, Olympia, Washington.
- Holden, T.J., Newton, Sylvestri, P. and Diaz, M. (1997). *How to Select Hazardous Waste Treatment Technologies for Soils and Sludges: Alternative, Innovative and Emerging Technologies*. Noyes Data Corporation, U.S.A, pp. 99 – 100.
- ISAAA Brief No. 8. Ithaca, N.Y.: International Service.
<http://www.bch.biodiv.org>)

- James, C. (1998). Global Review of Transgenic Crops: 1998.
- Jones, W.R. (1998). "Practical Applications of Marine Bioremediation." *Current Opinion in Biotechnology* 9: 300 – 304.
- Khosho, T. N. (1994b). *Conservation and Sustainable Utilization of Biodiversity in the Developing Countries: A Blueprint For Action*. Indian National Science Academy, Delhi.
- Khoshoo, T. N. (1996). "Biodiversity in the Developing Countries." In *Biodiversity, Science and Development: Towards a New Partnership*. (di Castri, F and Younnès, T., (eds). CAB INTERNATIONAL, Oxon OX10 8DE, UK.
- Layokun, S.K.; Solomon, B.O. and Ojo, A.O. (1988). "Some Growth Characteristics of Ife Cowpea." *Rhizobium* 9. JNSChE Vol. 7, No.1.
- Layokun, S.K.; Umoh E.F. and Solomon, B.O. (1987). "A Kinetic Model for the Degradation of Dodecane by *Pseudomonas Fluorescens* Isolated from the Oil Polluted area of Warri in Nigeria." *JNSChE Vol. 6. No.1*.
- Lee, K. and Levy, E. M. (1991). Bioremediation of Waxy Crude Oils Stranded on Low-energy Shorelines. *Proc. 1991 Oil Spill Conference*. American Petroleum Institute, Washington, D.C.
- Madras.P.326.
- May, 491-4.
- McNaughton, S.J., Stephen, J.R. Venosa, A.D. Davis, G.A. Chang, Y. and White, D.C. (1999). "Microbial Population Changes during Bioremediation of an Experimental Oil Spill." *Appl. Environ. Microbiol.* 65(8): 3522 – 3574.
- Medan, Indonesia. Oct. 20th –22nd 2000. Pp. 154.
- Mendelsohn, I.A., Hester, M.W. and Hill, J.M. (1993). "Effects of Oil Spills on Coastal Wetlands and their Recovery." *OCS Study MMS 93-0045*. US Department of the Interior, Minerals Management Service, Gulf of Mexico OCS Regional Office, New Orleans, La. 46p.
- Nkala, C.D. (2000). "Bioreactor Treatment of Lube Oil-contaminated Soil." *M. Sc. Thesis, University of Port Harcourt, Port Harcourt*.
- Nnubia, C. and Okpokwasili, G.C. (1993). "The Microbiology of Drill Mud Cuttings from a New Offshore Oilfield in Nigeria." *Environ. Pollut.* 82: 153 – 156.
- Nweke, C. O. and Okpokwasili, G.C. (2003). Drilling Fluid Base Oil Degradation Potential of a Soil *Staphylococcus* Species. *Afr. J. Biotechnol.* 2 (9): 1 – 6.
- Nwokoro, C.G. and Okpokwasili, G.C. (2003). "Bioreactor Treatment of Oil-contaminated Sediment." *Nig. J. Microbiol.* 17(2): (in press).
- Oboirien, B. O.; Amigon, B.; Ojumu, T. V.; Ogunkunle, O. A.; Adetunji, O. A.; Betiku, E. and Solomon, B. O. (2005). "Substrate Inhibition Kinetics

- of Phenol Degradation by *Pseudomonas Aeruginosa* and *Pseudomonas Fluorescence*." *Biotechnology*, 4 (1): 56-61.
- Ochsner, U.A., Reiser, J. Fiechter, A. and Witholt, A. (1995). "Production of *Pseudomonas Aeruginosa* Rhamnolipid Biosurfactants in Heterologous hosts." *Appl. Environ. Microbiol.* 61: 3503 – 3506.
- Okafor, Ndaka (2005). *Personal Contributions to a Blueprint for Sustainable Management of Nigerian Resources: Sustainable Bioresources Management and Biotechnology Intervention*. Raw Materials Research and Development Council, Abuja.
- Ojumu, T. V.; Bello, O. O.; Sonibare, J. A.; and Solomon, B. O. (2005). "Evaluation of Microbial Systems for Bioremediation of Petroleum Refinery Effluents in Nigeria." *African Journal of Biotechnology*, 4 (1): 31-35.
- Okpokwasili, G. C. (1994). Pollution Control: The Increasing Role of Bioremediation. *Biotechnology in National Development*, Proc. 1993 National Workshop, NASENI, Lagos. pp. 234 – 239.
- Okpokwasili, G.C. (1998). "Bioremediation: The Microbial Solution to Our Waste Management Problems." Lead Paper Presented at the 22nd Annual Conference of the Nigerian Society for Microbiology, FUT, Owerri.
- Okpokwasili, G.C. (2002). *Environmental Biotechnology Sectoral Working Document for the First National Biotechnology R & D Workshop*, NABDA, Abuja.
- Okpokwasili, G.C. and Nnubia, C. (1999). "Biodegradation of Drilling Fluids by Marine Bacteria from Below an Offshore Oil Rig." *J. Sci. Engr. Tech.* 6 (1): 1420 – 1428.
- Okpokwasili, G.C. and Oton, N. (2004). "Comparative Applications of the Bioreactor and Shake Flask Systems in the Laboratory Treatment of Refinery Sludge." *Seventh International Symposium on Geotechnology and Global Sustainable Development*, Helsinki, Finland.
- Okpokwasili, G.C. and Ibe, S.N. (1988). "Genetically Engineered Microbes and Oil Degradation." *The Petroleum Industry and the Nigerian Environment*, Proc. 1987 International Seminar. pp. 97 – 103. NNPC, Lagos.
- Okpokwasili, G.C. and James, W.A. (1995). "Microbial Contamination of Kerosene, Gasoline and Crude Oil and Their Spoilage Potentials." *Material und Organismen* 29: 147 – 156.
- Onyia, C. O. Uyub, A. M., Akunna, J. C. and Omar, A. K. M. (2000). "Bioaugmentation of Palm Oil Mill Effluent (POME), for Studies into Its Nitrification Activities." *Proceedings of the Indonesian-Malaysian-Thailand-Great Triangle (IMT-GT) International Conference on Land Use, Management and its Application*, 2000. p586
- Onyia, C. O., Akunna, J. C., Uyub, A. M., Norulaini, N. A. and Omar, A. K. M. (2001). "Enhancing the Fertilizer Value of Palm Oil Mill Sludge:

- Bioaugmentation in Nitrification." *Journal of Water Science and Technology*, 44: 10, 56-62.
- Onyia, C. O. (2002). "Biological Nitrogen Removal Processes from Industrial Waste Waters: The Case for Palm Oil Mill Effluent." Ph.D. Thesis, University Sains Malaysia, Malaysia.
- Roscoe, Y.L., McGill, W.B. Nyborg, M.P., and Toofood, J.A. (1989). "Method of Acceleration of Oil Decomposition in Soils." In *Proceedings of Workshop on Reclamation of Disturbed Land*. Northern Forest Research Centre, *Alberta Information Report* 462 – 470, Edmonton.
- Rosenberg, E. (1986). "Production of Surfactants to Facilitate Residual Oil Mobilization. *CRC Crit. Rev. Biotechnol.* 3: 109 – 132.
- Sharma M. (2004). India: Biotechnology Research and Development <http://www.cgiar.org/biotech/rep0100/sharma.pdf>. Accessed on 19th April, 2007.
- Singh, B.K. 2004, *Biodiversity Conservation and Management*. Mangaldeep Publishers, Jaipur
- Solomon, B.O., Fatile, I.A. and Layokun, S.K. (1986). "Selection of Microorganisms for Treatment of Oil Spillages and Hydrocarbon Effluents from Petroleum Industries: Maximum Specific Growth Rate, Yield and Maintenance Requirements." *JNSChE Vol. 6. No.1*.
- Swaminathan, M.S. (1992). "Biodiversity and Biotechnology." In: *Biodiversity Implication on Tetranitrate Reductase.* *Nature Biotechnology* 17(5)
- USEPA. (1991). Soil J.A. Vapour Extraction Technology. EPA/540/291/003. Office of Research and Development, Washington, D.C.
- USEPA. 1992. A Technology Assessment of Soil Vapour Extraction and Air Sparging. EPA/600/R-92-173. Office of Research and Development, Washington, D.C.
- WCMC (1992). *Global Biodiversity: Status of Earth's Living Resources*. World Conservation Monitoring Centre, London.