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INSTRUCTIVE LESSONS FROM THE EXCITING WORLD OF MICROBES

By

Gbolahan 'Ola Babalola
Professor of Microbiology

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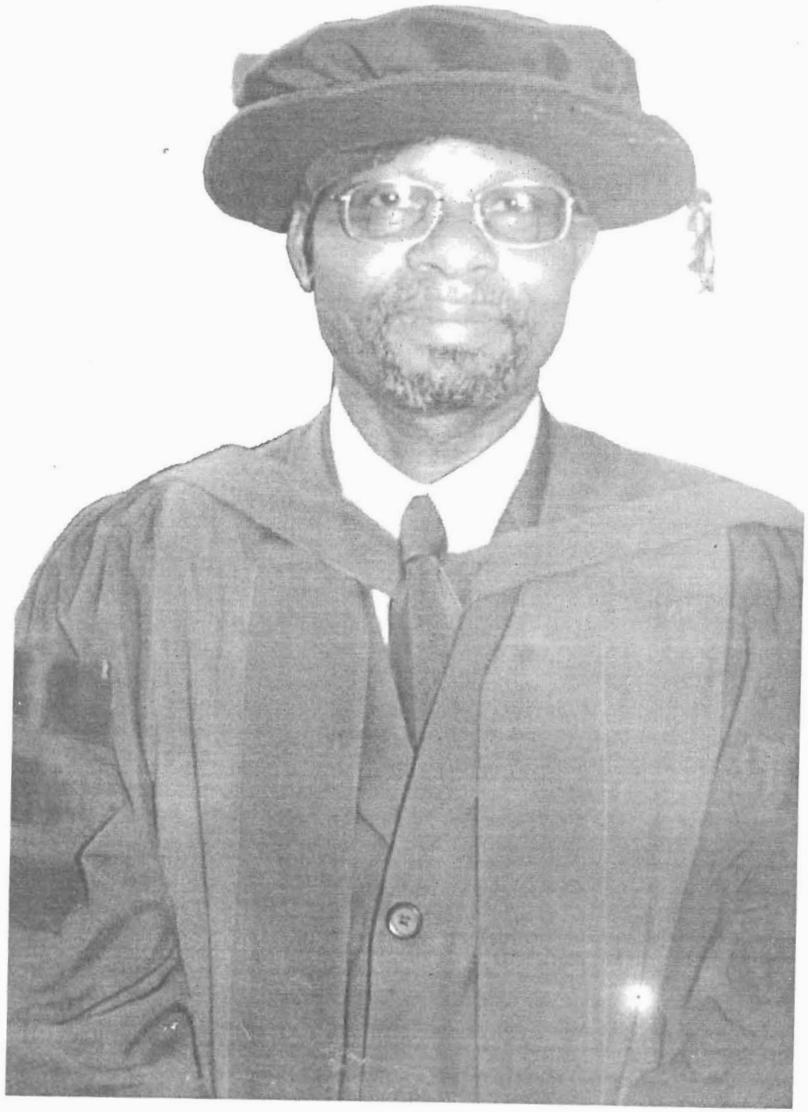

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An Inaugural Lecture Delivered at
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INTRODUCTION

The Vice Chancellor, distinguished ladies and gentlemen, it is my pleasure to be here today to give an account of my professional academic sojourn thus far. I am doing this with a mixed feeling of satisfaction of partial accomplishment of goals on one hand, and of disappointment on the other. In one sense, however, I am happy that I have been privileged to be the first fully home-brew Professor of the Department of Microbiology, being one of the pioneer students of the department. You will agree with me that this is a rare honour for which I am proud. The other side of my mixed feeling is that I am convinced that the system I have gone through in time and space hampered the full realization of my potential. Not in the sense that the "brew" was sour, but that it did not have the opportunity of the right environment needed to "mature (age)" in the cellar! In the fermentation and brewing parlance, best wines are those that are stored up to cure in the cellar before they are put on the table. This account of my academic voyage is, therefore, a summary of the modest contribution of my laboratory to knowledge, taking lessons from microorganisms!

MY PROFESSIONAL UP-BRINGING

The early stages of my career were not as direct as would normally have been expected. But this development turned out later to be my strength. My training cut across the major areas of microbiology. My undergraduate project was on environmental/water microbiology. For my M.Sc., I ventured into medical bacteriology. As a Graduate Assistant, I speculated into applied microbiology. I started the investigation into possible generation of biogas from the weed plant *Eupatorium odorata*. Meanwhile, there was (at that time) this need of a virologist in the department, and indeed everywhere. Virology was a curriculum discipline in which we had almost nil exposure. I became interested and decided to have my

PhD training in medical virology in the Medical School, University of Birmingham, England. The excitement was immense that on my return from England I came back with some equipment, courtesy of my department. I had worked on the search for a suitable vaccine against genital herpes, a sexually transmitted disease caused by herpes simplex virus type 2. It was then a major scourge as HIV/AIDS was just emerging into the scene. Quite naturally, my desire was to continue in my search for vaccines against viral diseases. But I think I qualified at the wrong time! Not only were there no facilities to complement the ones I brought for basic research which I was used to in my training, the international community was hostile to Nigeria because of the type of governance of the time. This made it difficult to obtain partnership support of any kind. The frustration was imminent requiring courageous and elastic patience, and real hard work. But with the wide scope and diverse background training that I had as specified earlier, I was able to adjust, make do with the facilities available in the university and personal sponsorship. So the road had not been smooth.

RESEARCH ACCOMPLISHMENT

My research activities have been broad-based, basic and applied. However, the focus was on the significance and efficiency of "smallness" as embodied in microorganisms. The general perception of microorganisms by us has generally been a negative one, even among the educated individuals. But I must submit here that this concept, even though partially true, is most often exaggerated. Indeed, there is a lot to learn from the world of microorganisms. Like all living things, they seek to compete for available resources to guarantee their survival. However, their versatility and the relative greater efficiency of cellular organization accord them a higher rate of success. This is exemplified in a tremendous array of strategies with which they take command of

their environment and hence survival. Thus, their superior and efficient survival strategy endows them with their threatening composure over other organisms. Consequently, we tend to see them generally as our enemies. But we now know better that we have every cause to be grateful to microorganisms for without them, other lives on earth may have gone into extinction. Indeed, most of the conceived negative impacts of microorganisms are what we actually need to exploit to our advantage and for our survival. We surely have more benefits to derive from their activities than otherwise.

My laboratory has been examining these aspects of microorganisms that pose actual negative impacts on us, and how to abate them. We have also been looking at the exploitable ones for our benefits. Thus, we recognize the need to address both the undesirable and beneficial aspects of the activities of microorganisms.

UNDESIRABLE ACTIVITIES

Microbial transformation of drugs: In collaboration with colleagues (the late Prof. Francis Ogungbamila and Prof. S.A. Adesanya) in the Faculty of Pharmacy, our studies revealed that some of the adverse toxic effects of drugs, might have been caused by microbial transformation of the main components (including syrups and other binding agents that provide good nourishment for the growth of microorganisms) of the drug into some toxic or inactive derivatives. For example, we have found out that some air-borne fungal species are capable of converting the analgesic paracetamol (*p*-Acetaminophenol) into some derivative metabolites one of which is suspected could be toxic. The suspect derivative is 4-aminophenol, which if it accumulates to a high concentration in tissues could cause fatality. It impacts pink to dark colouration to the drug depending on the concentration.

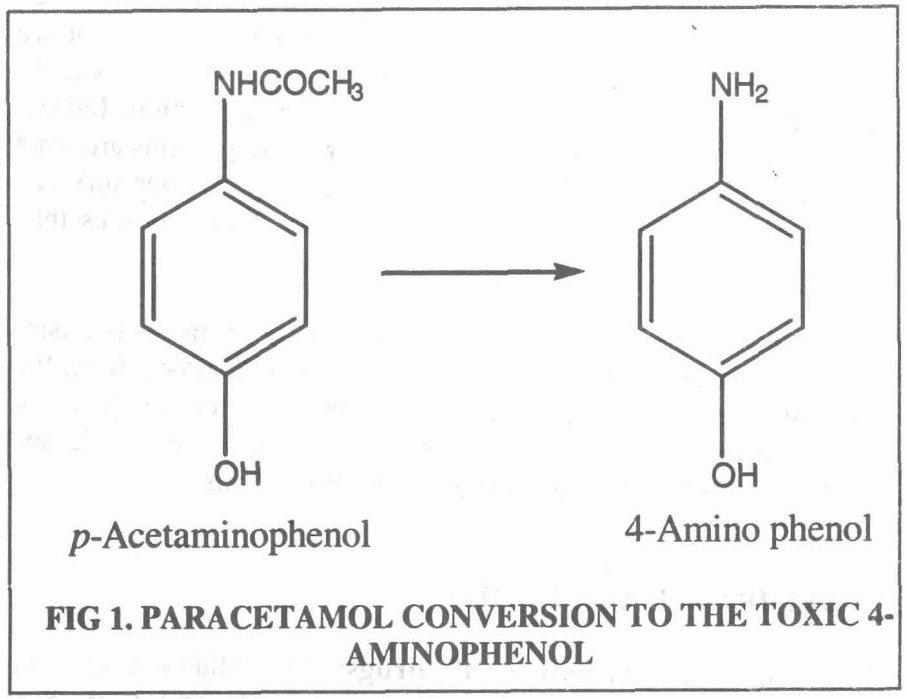

FIG 1. PARACETAMOL CONVERSION TO THE TOXIC 4-AMINOPHENOL

There is, therefore, the need to exercise caution in the process of production and choice of the method of storage of drugs to avoid contamination, and hence inactivation of their therapeutic effect by microorganisms. This should not be too difficult to accomplish by proper implementation of sanitary and sterilization guidelines during production, adequate storage conditions, and careful dispensing and handling.

However, I must draw your attention to the fact that our knowledge of the metabolic activities of microorganisms on drugs has been very useful in our understanding of the metabolic pathways of drugs in animals. The toxicity of the β-gentiobioside, which occurs in the

seeds of bitter almond, was assigned to its conversion to cyanide by intestinal bacteria after oral administration. A dysfunction of the blood (dyscrasias) associated with the use of chloramphenicol has also been linked to the product of its metabolisms by intestinal bacteria (Schelein, 1973). In many ways, the microbial metabolic pathways are essentially similar to what obtains in animals. In our study, accumulation of the 4-aminophenol affected the growth of the microorganisms, hence the suspicion that it could also be toxic to man. Indeed, it has been implicated in renal and other tissue toxicities (Hegedus and Nayak, 1991). Consequently, microorganisms are very useful research tools in this regard.

Combating microbial infection and contamination: It is common knowledge that some microorganisms constitute nuisances to crops by infecting their seeds and seedlings, thus creating socio-economic problems. Usually, this is accompanied by huge losses of yield of food crops such as cowpea, beans, tomatoes, pepper, and maize among others. Some other microorganisms also decompose foods and other biological products, and biodegrade non-biological substances such as crude oil and its by-products. Some cause diseases of animals too. We have attempted to address some of these negative effects as follows:

1. **Application of the anti-microbial effect of *Vernonia amygdalina* (bitter leaf):** We have tried successfully, the use of the extract of the leaves of a local plant *Vernonia amygdalina* (bitter leaf) as a substitute for hop in the brewing of larger beer, for the purpose of imparting a bitter taste as well as serving as a preservative. Our product passed the organoleptic (taste) test, and was comparable to the existing products in the market (Table 1). The larger beer produced also survived a shelf life of more than a year.

Table 1: Comparative properties of sorghum beer flavoured with 1.2 g liter^{-1} bitter leaf extract (BLE) and commercial beer

Analysis	BLE Beer	Commercial Beer
Final Gravity (% w/v)	2.06	1.50
Real Extract	4.00	3.50
Calorific Value	37.14	41.16
Refractive Index	38.00	35.00
pH	4.02	3.50
Original Gravity (% w/v)	11.04	11.36
Alcohol Content (% v/v)	3.02	3.88
Colour (°EBC)	8.20	7.20

A preparation of the extract using a local cream (ori) as a carrier was also tested successfully for topical application in the treatment of wound against bacterial infection in mice. For lack of fund, we could not carry on further to a standard research conclusion through detailed animal test experiments. It is very expensive to maintain an animal house needed for such experiments. Going by the awful low level of funds available as research grant, we simply could not afford it even up till today. This plant has a huge potential for use as a source of antibacterial preservative/therapeutic agent without fear of risk of side effects as it is a well known food vegetable in the southern parts of Nigeria, commonly called "Ewuro" or "Bitter-leaf".

2. **Anti-microbial active compounds from *Pycnanthus angolensis* (Warb):** In another search for anti-microbial agents from natural sources, a collaborative study with Prof. S.A. Adesanya revealed the potential of the extract of the wood of *Pycnanthus angolensis* **(Warb).** The

wood is used locally as chewing sticks. The extract of the heartwood of the tree produced a new isoflavone 7,4'-dimethoxy-2'-hydroxyisoflavone in addition to the known 2'-hydroxyformononetin. The new isoflavone was active against some test bacteria and fungi, and the minimum inhibitory concentration (MIC) ranged between 350 µg ml^{-1} and 360 µg ml^{-1}. Meanwhile, Simon *et al.,* of the Rutgers, The State University of New Jersey, New Brunswick, NJ, USA) have reported other extracts of the plant (sargaquinoic acid, sargachromenol, and sargahydroquinoic acid) as showing antioxidant and anti-inflammatory activities. The compounds were reported to effect the reduction of generation of nitric oxide synthase and cyclooxygenase-2. They filed in their findings for patent rights on May 5, 2003.

It is disheartening that we could not carry further our investigation on the potential of this plant basically because the research environment here has not been conducive over the years. We first observed the potential, and identified one of the active principles of the plant (Babalola, 1988; Omobuwajo *et al.,* 1991), only for some others to claim the credit, albeit legally. What a loss of glory to our university, and Nigeria! Surely there is a lesson to learn from this experience which I am sure is not isolated and hence is peculiar to the poor handling of our educational system. Perhaps there is also another important lesson to learn here. The antioxidant mechanism of action of the extracts from this plant as stated above relies on their ability to scavenge and/or prevent excess of free radicals in the system (Elad, 1992; Dalton, 1995; Gala and Abdou, 1996), and hence the submission that the compounds should be useful for the treatment of a variety of illnesses caused by free radicals and inflammatory factors (including cancer and diabetes) in human, animals and birds. In this regard, there should be

some logic in the claims of our native long-established healers that some of our herbs could cure an array of human illnesses. Such herbs are conventionally referred to by the Nigerian Yoruba ethnic group as "gbogbo ise" i.e. "heal all". I want to submit here that there is the need to consider a much more dedicated and coordinated research into what is left of our traditional therapeutic remedies, rather than our usual scornful relegation of our rich cultural heritage and native knowledge into the background. Else, someone from the "developed" world would soon come up as usual to report some remedies for HIV/AIDS and the like, when the original source is from what we already have in our indigenous "Pharmacopoeia", merely waiting to be refined and explained!

3. **Anti-microbial activity of synthetic aromatic chlorochromates:** Because of the versatile nature of microorganisms, the search for synthetic anti-microbial agents must continue. We have examined the anti-microbial potentials of two synthetic aromatic compounds, quinolinium chlorochromate and pyridinium chlorochromate. The later is used primarily in chemistry as an efficient reagent for oxidation of primary and secondary alcohols to carbonyl compounds, and can be synthesized in the laboratory. The two compounds were prepared and supplied by Professor Craig Obafemi of the Department of Chemistry with 98 % purity. However, pyridinium chlorochromate is also available commercially as a chemical reagent. The MIC of the compounds against the test bacteria used were in the range 125-250 $\mu g\ ml^{-1}$ and 250-500 $\mu g\ ml^{-1}$ for the pyridinium and quinolinium chlorochromates respectively, while those of the parent compounds, pyridine and quinoline, were 4000-5000 $\mu g\ ml^{-1}$ and >2500 $\mu g\ ml^{-1}$, respectively. The MIC of perfloxacin, a drug which has

been reported as effective for the treatment of bacterial infection, was 125-250 $\mu g\ ml^{-1}$ the same as for pyridinium chlorochromate.

These compounds have exhibited effective activity against simulated bacterial infection in mice by topical application in the treatment of *Pseudomonas aeruginosa* infection of wound, and subcutaneous application in the treatment of systemic *Staphylococcus aureus* infection. In one experiment, the administration of approximately 22.0 $\mu g\ g^{-1(body\ weight)}$ of pyridinium chlorochromate and 45 $\mu g\ g^{-1(body\ weight)}$ of quinolinium chlorochromate to infected mice reduced mortality by 50 % and 67 % respectively when compared with the untreated controls (Table 2). However, higher mortality rates were recorded at higher doses, but were more pronounced in the infected (100% for pyridinium, 83% for quinolinium) than uninfected but treated mice (83% for pyridinium, 67% for quinolinium). Considering our observation in the toxicity test, it would appear that quinolinium chlorochromate was less toxic than the pyridinium chlorochromate. The estimated LD_{50} (50 % lethal dose) was approximately 76 $\mu g\ g^{-1}$ and 33 $\mu g\ g^{-1}$ respectively. Pyridinium chlorochromate was, therefore, more active than its quinolinium counterpart *in vitro*, but was more toxic to the tissue and less effective *in vivo*. The observed effective treatment dosage-levels recorded (22 $\mu g\ g^{-1\ (body\ weight)}$ of pyridinium chlorochromate and 45 $\mu g\ g^{-1}$ $^{(body\ weight)}$ of quinolinium chlorochromate) were comparable to the 50 mg $kg^{-1\ (body\ weight)}$ treatment of experimental endocarditis caused by *Staph. aureus* using a similar compound ciprofloxacin in rabbits (Carpenter *et al*, 1986). However, the two compounds exhibited antagonistic effect against each other when tested combined in *in vitro* experiments. It is conceivable that the absolute levels of

efficacy of the compounds should be much higher if further purified. Further pharmacological studies should establish their suitability for therapeutic applications against bacterial infections in man and animals.

Table 2: Effect of pyridinium and quinolinium chlorochromates treatments on experimental *Staphylococcus aureus* infection in mice*.

	Mortality at 12 h post treatment‡														
	Pyridinium chlorochromate						**Quinolinium chlorochromate**								
Dose	Infected			Uninfected			Dose	Infected			Uninfected				
($\mu g\ g^{-1}$)	1st	2nd	3rd	1st	2nd	3rd	($\mu g\ g^{-1}$)	1st	2nd	3rd	1st	2nd	3rd		
44.60	0	2	6	0	1	5	90.00	0	1	6	0	2	4		
22.14	0	1	2	0	0	2	45.00	0	1	1	0	0	2		
Saline	0	2	4	0	0	0	Saline	0	1	3	0	0	0		

* The mice were in-bred and there were six mice in each treatment group.

‡ Treatment was administered thrice (1st, 2nd, 3rd) at each dosage level at 12 hourly intervals

The compounds have also shown tremendous potential in the application for seed protection, seed disinfection and seed disinfestation. Seed treatment is an important procedure in agriculture for protective storage of harvest, and improved crop yield among other benefits. For instance, the average yield of cowpea in West Africa estimated at 100 – 300 kg ha^{-1} is still far from expectation (Kachara *et al.*, 1988). Barring any adverse effect, the net yield potential has been estimated at 2000 kg ha^{-1}, and the world average yield is 337 kg ha^{-1}. Studies have shown that grain yield loss from bacterial blight due to infection of

cowpea by different strains of *Xanthomonas campestris* (the major pathogen of the seedlings) could be as high as 67 %. In our experimental pre-planting treatment of infected cowpea seeds (simulated with a strain of *Xanthomonas campestris* –strain IT97K-1069-3, isolated from a farm in Ikenne, Ogun State, Nigeria and supplied by IITA (International Institute of Tropical Agriculture, Ibadan, Nigeria)), pyridinium chlorochromate reduced infection of seedlings significantly. The germination capacity, root and shoot lengths, and vigour index of the treated infected seeds were significantly better than those of the infected but untreated seeds, and compared favourably with the uninfected/untreated control (Plate 1). At the concentrations used (100 – 300 µg/ml), the treatment did not have any significant adverse effect on nodulation, an essential feature of cowpea in a symbiotic natural fertilization of the soil. The treated plants were generally comparable to those that were not infected and not treated. Similar results were obtained for maize, pepper and tomato, where the treated seeds survived simulated infections by corresponding bacterial pathogens.

With the excellent tolerance of the test animals and seeds for the chemical treatment at the concentration levels which reduced the risk of bacterial infections, it is reasonable to conclude that these compounds have a potential for use as prophylactic or chemotherapeutic anti-microbial agents in plants and animals.

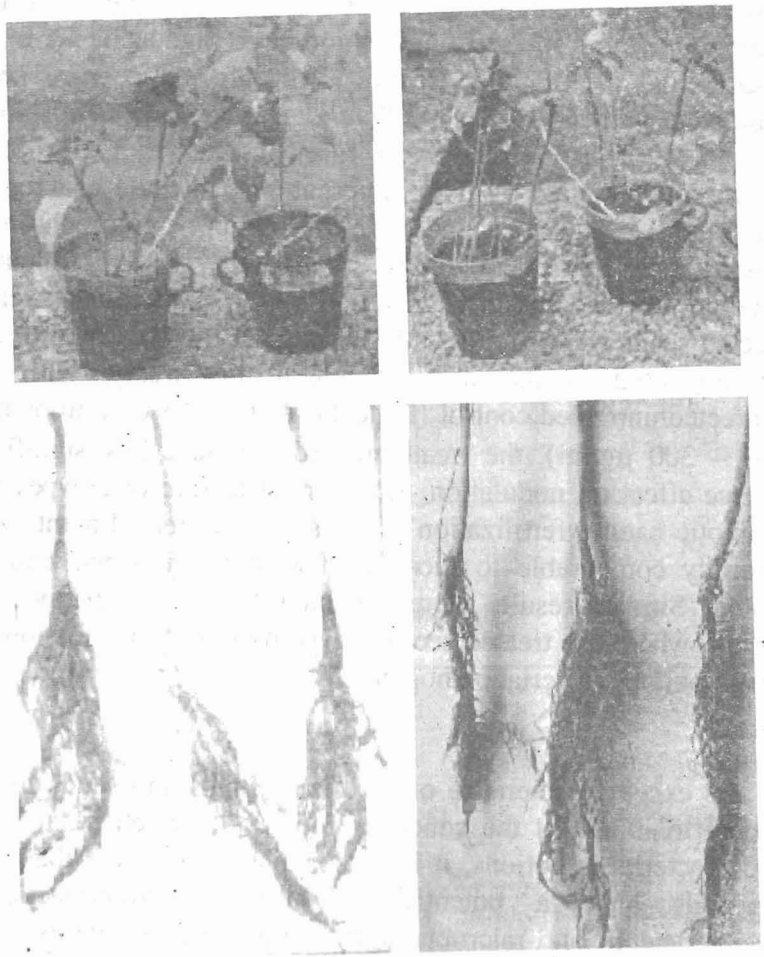

Plate 1: The shoots (Top) and the roots (Bottom) of infected but treated with pyridinium chlorochromate (100 µg/ml), (Left) and infected but untreated (Right) cowpea plants. Infection was simulated with a strain of *Xanthomonas campestris* isolated and supplied by IITA, Ibadan, Nigeria.

MICROORGANISMS AND THE ECOSYSTEM

As we feel concerned about the undesirable effects of microbial activities and look for ways of combating them, it is much more imperative that we consider the important roles of microorganisms in the stabilization of the ecosystem. In this regard, it is very important that we examine how our activities impact the positive contributions of microorganisms to the ecosystem and hence our survival, and check them. In collaboration with Professor A. Olayinka of the Department of Soil Science, our studies revealed a potential adverse effect of accumulation of copper in the soil on the microorganisms that are crucial to soil chemistry, and hence agricultural productivity of the soil. To be specific, we have observed that accumulation of copper in the soil caused a significant reduction in the activities of nitrifiers. Nitrifiers are microorganisms that are involved in the conversion of inorganic nitrogen compounds in the soil into forms that are readily assimilated by plants. In this way, the availability of the nitrogen in the food chain is ensured. As we all know, nitrogen is one of the most essential elements of life in all organisms; plants and animals, big or small (carbon, hydrogen, phosphorus and sulphur are the others), constituting an important component of biological macromolecules, the proteins and nucleic acids.

Further investigations in my laboratory revealed that accumulation of copper in the soil significantly affected symbiotic biological fixation of atmospheric nitrogen in the soil. The percentage germination, average number of nodules, and average diameter of nodules of cowpea reduced as the concentration of copper sulphate increased, by approximately 25 %, 39 %, and 75 % respectively at 300 µg/ml concentration of the solution (Table 3).

and the leguminous plants e.g. alfalfa, clover, beans and peas, forming nodules in the roots of the plants. It is a natural process of fertilizing the soil, thereby replenishing used or lost nitrogen content of the soil. It is estimated that a properly cultivated cowpea plantation could fix about130 Ib nitrogen per acre by this symbiotic relationship (Fery and Singh, 1997) equivalent to 50-300 kg urea fertilizer per hectare.

Table 3: Effect of Copper Sulphate on the Development and Nodulation of Cowpea.

TREATMENT WITH COPPER SULPHATE	GERMINATION (%)	AVERAGE NO OF NODULES	AVERAGE DIAMETER OF NODULES (cm)
Untreated control	85	62(6.1)	0.4(0.1)
100 µg/ml	71	53 (3.0)	0.3(0.1)
200 µg/ml	71	47(3.7)	0.2(0.1)
300 µg/ml	62	38(0.6)	0.1(0.0)

Figures in parentheses are the standard deviations from the means

In general, about 140 x 10^{12} g of atmospheric nitrogen is biologically fixed per hectare of land per year globally, about double the amount fixed non-biologically (industrial, combustion or lightning) (Table 4). More than half of the biological fixation is accounted for by agricultural land use. You will agree with me that there is the need to look critically into the use of copper sulphate in the treatment of fungal infection of cocoa and other plants for the purpose of improving the quality of yield. We must, make conscious efforts to educate ourselves to avoid indiscriminate use of

copper sulphate as an anti-microbial agent in agriculture else the hidden losses from the application of copper sulphate as an anti-microbial agent may outweigh the expected gains.

Table 4: Estimates of the Amount of Nitrogen Fixed on a Global Scale.

Type of fixation	N_2 fixed (10^{12} g per year, or 10^6 metric tons per year)
Non-biological	
Industrial	about 50
Combustion	about 20
Lightning	about 10
Total	**about 80**
Biological	
Agricultural land	**about 90**
Forest and non-agricultural land	**about 50**
Sea	about 35
Total	**about 175**

Data from various sources, compiled by DF Bezdicek & AC Kennedy, in Microorganisms in Action (eds. JM Lynch & JE Hobbie). Blackwell Scientific Publications 1998.

EXPLOITABLE ACTIVITIES OF MICROORGANISMS

I mentioned earlier on that our perception of microorganisms as "our destructive enemies" of economic and health development is

in the real sense far from the whole truth. For a given metabolic or

chemical reaction that is possible, such a reaction is comparatively executed with greater efficiency in microorganisms in terms of simplicity, sometimes speed and accuracy, than you will find in higher organisms or industrial processes. For instance, the biological nitrogen fixation process mentioned earlier is also possible by an industrial process where nitrogen gas is reacted with hydrogen gas under very high pressure and temperature to create ammonium. However, the cost of production is highly prohibitive, requiring large amounts of fossil fuels to provide the hydrogen gas required in the reaction. Even where a microbial system is slow, it has been programmed to be so by the self-regulating systems of microorganisms for maximum utilization of resources, and such restrictions can be removed by laboratory manipulation techniques in consideration for industrial or adaptive applications. This comparably higher efficiency of the microbial system is inherent in the simple nature, short generation time (rate of turn-over) and versatility of the microorganisms. They are basically miniaturized bioreactors possessing technological behaviour.

Over the years, human beings have resorted to solving some of our problems by reaching out to explore and adapt the various activities and technological behaviour of microorganisms perceived as "good" or "bad" for the benefit of mankind. Various examples include the use of microorganisms for industrial manufacturing processes (amino acids, antibiotics, vitamins, enzymes), applications in food processing (single cell protein, food and alcoholic beverages fermentation), and adaptation of the knowledge of infectious diseases for preventive intervention strategies such as in immunization regimes (eradication of small pox and the successes in tetanus, diphtheria and polio diseases should easily come to our mind). The latest is the application of gene manipulation technique (popularly known as genetic engineering) for solving various industrial, agricultural, environmental and medical problems. The academic field is generally referred to as

"Biotechnology" and the processes and tools used are based on our knowledge of the microbial systems.

In our modest ways, my laboratory has tried to look at some possible adaptations of microbial activities for solving some problems:

Cottage-type recycling of household wastes. The process involves an age-long natural activity of microorganisms in recycling elements, and constitutes an important component of what is generally referred to as "biogeochemical cycle". The process is called "composting", but in this natural way it is slow to satisfy the volume of needs. Our focus is to evolve a semi-mechanized process line that will speed up the rate of output several-fold. The product (compost) is suitable for use as an organic fertilizer and/or soil conditioner. Used as a soil conditioner, it is expected to improve the porosity of clay soil and water-retaining capacity of sandy soil.

In partnership with Professor Olu Odeyemi of my department, a project commissioned by Shell Petroleum Development Company, Warri Branch, was executed to test our idea. I must say here that there are highly mechanized approaches being practised in industrialized countries today but we have chosen a cottage-type approach because we believe that some technologies should be adaptable to suit local needs in sustainable and yet cost effective ways. It is of no use importing a technology that is either unmanageable in terms of manpower and maintenance, or is unsustainable in terms of cost. Drawing a lesson from microorganisms, small-scale processes also have their advantages, including adaptation for decentralization and reduction to a less technical and manageable scale. Besides, our approach has

additional advantages of an all-the-year round operation (weather-

proof) as well as providing for the avoidance of the risk of groundwater pollution that is attendant with the open-air high-tech methods.

Our processing plant consists mainly of a refuse collecting depot; a sorting point; the shredding depot; the composting shed, and the maturing and packaging depot. All of these occupied an area of about 3 acres of land. Approximately 203 tonnes of household refuse was collected in a period of one year of the project. About 134 tonnes (66%) of the refuse was finally sorted out from the lot and shredded (pulverized) to obtain about 60% volume reduction. Shredding is also meant to increase the surface area for microbial activity. Compost mix was constituted in batches employing the windrow method. Each batch was a mix of shredded refuse added to bulking agents and microbial inoculants making up an average wet weight of 10 metric tonnes. The project generated more than 75 metric tonnes of mature fairly dry compost by this process.

Each composting process lasted approximately 5-6 weeks. Samples of the raw materials (shredded refuse, and other agents used) and finished product (compost) were taken for laboratory analyses. Samples of the composting mixture were also taken at time intervals for laboratory analyses. Representative samples of mature (cured) compost were also sent to IITA, Ibadan, Department of Agronomy, University of Ibadan (UNIBADAN) and Obafemi Awolowo University (OAU), Ile-Ife, for independent peer laboratory analyses and field applications, and assessment as a soil conditioner/manure. The results of the laboratory analyses (Table 5) showed that the major (C, N, P and K) and minor (Mg, Fe

TABLE 5: Comparative Values of Physical, Chemical and Biological Parameters for the Raw Materials (refuse and cow-dung), the Compost Mix and Mature Compost. The Figures Given are Averages for Eight Different Batches of Compost Processing.

PARAMETERS	REFUSE	COW DUNG	COMPOST MIX	MATURE COMPOST
PHYSICAL FACTORS				
PH	5.12	8.66	5.67	9.7 (8.9)*
Moisture (%)	70.00	79.67	74.40	20.0 (15-25)
Ash Content (%)	20.40	28.60	29.60	49.9
MAJOR ELEMENTS				
C (%)	43.63	39.70	39.50	27.8
N (%)	0.97	0.85	1.13	1.13 (10-15)
P (%)	0.30	0.46	0.59	0.84 (15-20)
K (%)	0.85	0.67	1.22	1.08 (15-20)
TRACE ELEMENTS				
Ca (%)	2.43	1.67	5.07	3.2 (3-7)
Mg (%)	2.82	3.32	4.92	6.2 (0.5-1)
Fe (%)	0.77	0.00	1.53	1.5 (0.01-0.05)
HEAVY ELEMENTS				
Pb (%)	1.37	0.67	5.63	3.7 (<0.001)
Cu (%)	0.67	0.53	1.73	0.8 (<0.003)
Cr (%)	0.73	0.43	4.73	5.5 (<0.01)
MICROBIOLOGY				
Heterotrophic Bacteria (cfu/ml)	675	1991	583	258
Coliform (MPN 100^{-1}	38	44	31	0.0009 (0)
Fungi count (cfu/ml)	3910	3375	2900	295 (variable)

*** Figures in parentheses are recommended values.**

and Ca) elements of agronomic values were in appreciable quantities in the mature compost. Also, the heavy elements of health concern (Cu, Pb and Cr) decreased generally and to acceptable levels, as compared to the raw materials. The moisture

content and pH were also within acceptable limits. The main biological index of health importance i.e. the faecal coliform population was nil in the final product.

The results of field application from IITA, UNIBADAN and Obafemi Awolowo University, Ile-Ife showed very clearly that the mature compost produced in this project improved the agricultural productivity of the soil for the various crops (maize, cowpea and cassava), and compared favourably with commercial fertilizer in the greenhouse and field experiments. This was reflected in the improved growth rate and structural development, higher crop yield, as well as in the improvement of the nutrient content (especially N) of the crops.

With these findings, we are convinced and pleased that we have successfully demonstrated a sustainable and cost-effective process for the management of household refuse and its conversion to a product, which is safe and profitable for agronomic application. The process is less technical once it is put in place and hence it can be adapted for both rural and urban use. Our plant can also be adapted for integration with fish farming, using seepage from the composting process (rich in urea and nutrients) and other process by-products that can serve pond pH regulator and as food for the fish.

I must add here that if our method is finally perfected, it should be possible in the end to institute an integrated procedure for better management of domestic and some office solid wastes, which are creating a menace to health and aesthetics of the environment. At the moment, solid wastes are disposed by landfills and subsequent incineration that can produce other problems that might even be worse than the one intended to solve. Dioxins "dibenzodioxins" is a catchall term for compounds with two benzene rings bridged by two oxygen atoms. The polychlorinated derivatives have been

implicated as possible agents of cancer and disruption of the immune system. Incineration is a major source of these compounds and burning of domestic wastes is estimated to contribute about 30 % worldwide (Table 6). Besides, the landfill system is a potential source of groundwater pollution. Accelerated composting, therefore, should provide a relief in reducing these impacts on the environment.

Table 6: Estimated Average Annual Worldwide Emission Sources of Polychlorinated Dibenzodioxins (Pcdds) and Polychlorinated Dibenzofurans (Pdcfs).

SOURCE	KILOGRAMS/YEAR
Municipal Waste Incineration	1130
Biomass Combustion	350
Iron Metals Production	350
Cement Kilns (burning hazardous waste)	680
Cement Kilns (no hazardous waste)	320
Secondary Copper Smelting	78
Medical Waste Incineration	84
Unleaded Fuel Combustion	1
Leaded Fuel Combustion	11
TOTAL	**3004**

(Brzuzy and Hites, 1996)

A prototype composting "vessel" is being designed, constructed, and tested in collaboration with Engineer Dele Sanni of the Department of Agricultural Engineering, Obafemi Awolowo

University, Ile-Ife for this purpose. I am looking forward with a high optimism to a successful scale-up process in the nearest future, hoping that our efforts to get a grant for the project will be positive. Indeed, the benefits of forcing an improvement on the proper management of wastes is possible and desirable, making composting of wastes an avenue for making wealth from "trash"!

2. **Converting cassava wastes to wealth:** Cassava is an important plant source for some most staple foods in Nigeria cutting across the southern part of the country. Indeed, cassava is a common source of food in Africa and other developing countries of the world. Besides, cassava has been considered as a relatively cheaper source of industrial starch. Nigeria is currently promoting the planting of the crop for export. The implication is that a lot of wastes especially from the peels are expected. I have been looking into this problem with the view to transforming the "mess" into a profitable venture. In this regard, my laboratory has developed some ways of possible adaptations for an integrated cassava-processing factory for optimum utility of the crop. In this proposal, the central cylinder of the root tuber will be processed for either starch extraction and/or "gari" production among other possible utilization outlets, while the peels (wastes) shall be processed for the following products:

A. Microbiological culture medium: Microbiological media (food for micro-organisms) are generally made from plants and animal materials (potato, carrot, rice, hay, meat among others). This makes some sense since microorganisms commonly infect or contaminate these materials leading to what we perceive as spoilage. The microorganisms are simply looking for food to eat! In my laboratory, we have processed the extract of cassava peels for the production of liquid and solid microbiological culture media (the solid form contained the commercial solidifying agent, agar). The cassava peel broth sustained the

cultivation of isolates of bacteria comparable to the commonly used nutrient broth (Table 7). In the same vein, the solid preparation supported the growth and isolation of bacteria and fungi, including direct differentiation of starch-degrading ones from the rest. Although the counts were generally higher for the

Table 7: Total Colony Counts (Cfu) of Bacteria on the Cassava Peel Extract Agar and Starch Agar Media Seeded with Aqueous Samples from Different Sources.

	Cassava peel extract agar			Starch agar*		
Source of sample	**SH**	**PSH**	**NSH**	**SH**	**PSH**	**NSH**
SEWAGE	60	21	33	186	32	60
WELL WATER	76	44	36	105	34	21
STREAM WATER	10	33	20	18	84	30
DRAINAGE	94	35	22	96	78	40
SOIL A	82	15	24	74	36	39
SOIL B	38	23	20	27	61	33

*Soluble starch (1% w/v) was added as the main source of carbon supplement to the medium containing 0.3%(w/v) Bacto-beef extract. The counts are averages of duplicated plates. Plates were flooded with iodine after 36 h of incubation, following which colonies were identified as SH(starch hydrolysis), PSH(partial starch hydrolysis) and NSH(no starch hydrolysis).

commercial medium, the detection of colonial types was the same. In this way, the medium serves as a differential medium and provides for a reduction in cost and time. At the moment, a 500 g of imported commercial nutrient broth costs more than N

10,000.00. While the final formulation is being perfected, I am convinced the procedure is ripe for incubation and consequent adaptation for commercial production.

B. Microbial enrichment of cassava peel for livestock feeds:

Fermentation is a physiological process of some micro-organisms by which carbohydrates are metabolised producing intermediate products useful in the synthesis of other organic molecules that are essential for growth. Fermented foods are thus in many cases enriched with proteins, amino acids, vitamins among others. My laboratory has demonstrated the microbial enrichment of the spent marsh obtained in the production of cassava peel extract culture medium explained above. I am aware that sun-dried raw cassava and its peels have been used to raise livestock especially goats in the rural areas. Our approach here is to bring about an improvement on the nutritional value of the peels thereby maximizing its use in the production of animal protein. We have tried different consortia of microorganisms and obtained encouraging results as shown by analyses of our products that revealed substantial increases in the protein and vitamin contents, still leaving a high level of carbohydrate for the generation energy molecule adenosine tri-phosphate (ATP) (Table 8). Furthermore, preliminary studies on applications in animal experiments also showed positive results. Broiler chicks fed on a commercial feed supplemented with the microbial-enriched cassava peels marsh (20 %) gained more weight than the ones fed with the feed only (Table 9).

If the aforementioned experimental findings can be transformed into large-scale processes, you will agree with me that an integrated cassava-processing factory for optimum utilization of the root tuber is feasible. In the end, there should be an enhanced and cost-effective procedure for the processing of the crop for economic

benefits, providing for additional job opportunities and wealth creation. But perhaps the most important benefit should be in the contribution to reducing the environmental impact of inadequate management of cassava processing wastes, and turning it instead into wealth.

Table 8: Nutritive Contents of the Cassava Peel Marsh before (A) and after Enrichment with Some of the Different Consortia (B, C, D) Of Microorganisms Tested.

	ANALYSES (%)						
Batches	Crude Protein	Total Ash	Ether Extract	Crude Fibre	Total Carbohydrate	Total Nitrogen	Digestible Carbohydrate
A	6.13 (0.44)	5.42 (0.26)	0.78 (0.42)	11.93 (1.22)	80.68 (1.66)	9.80 (0.70)	68.75 (2.58)
B	9.23 (1.04)	7.84 (0.83)	0.56 (0.33)	14.56 (2.26)	80.47 (4.03)	14.73 (1.63)	72.43 (4.22)
C	10.70 (2.19)	7.94 (1.08)	1.40 (0.24)	14.22 (3.39)	78.75 (5.63)	17.09 (3.51)	73.97 (7.00)
D	10.66 (2.07)	8.05 (0.61)	1.34 (0.05)	14.32 (3.09)	77.46 (9.19)	17.10 (3.28)	75.83 (8.03)

Figures are averages and those in parentheses are standard deviations

Table 9: Comparative Weights of Two-Week-Old Broilers Fed Exclusively on a Commercial Feed (A) and those Fed on Commercial Feed Supplemented with Microbe-Enriched Cassava Peels Marsh (20 %)

	BROILER WEIGHT (G)	
SERIAL NUMBER	A	B
1	260.5	265.5
2	261.5	266.5
3	264.5	281.5
4	276.5	287.5
5	310.5	264.5
6	296.5	325.5
7	288.0	294.5
8	282.5	300.5
9	317.5	307.5
10	291.5	334.0
AVERAGE	284.95	292.75
STD DEVIATION	19.84	24.58

3. **Bioremediation of organic pollutants:** Earlier on I mentioned the ability of microorganisms to breakdown drugs. In the same token, some microorganisms do degrade organic pollutants from oil and other industries where heavy machines and organic chemicals are used. Such pollutants are conventionally treated by burning or using solvents/dispersants. In most cases, these methods do not remove the problems completely hence they are mainly palliative measures. Microorganisms are needed to completely remove the undesirable impact by breaking down repulsive pollutants into non-polluting forms. The use of microorganisms in

bioremediation/augmentation of pollutant treatments is becoming increasingly acceptable as an alternative to burning and chemical treatments. Dr. A.I. Okoh and Mr. A. Olaniran (currently on his PhD studies) have been working with me in the development of suitable microbial consortia for this process. They are both currently in South Africa carrying out more studies on some of the organisms we have isolated in Nigeria. Dr. Okoh has been working on the petroleum-degrading organisms, while Mr. Olaniran has been working on organochloride-degrading microorganisms.

TRANSFORMATION OF LESSONS LEARNT FROM RESEARCH

From the foregoing, there are clearly priceless lessons to learn from microorganisms, and these lessons are endless and instructive. Most often, they are adaptable for our socio-economic advancements. Some of the lessons are also informative for the sustenance of biologically diverse gene pool and hence the ecosystem of which human is a part. But the question remains as to why we have not made the best of our research experience and output for solving our various problems, especially for the advancement of our socio-economic and technological needs? Why do we always rely on finished technologies rather than developing some with local contents and for local needs? Put in the parlance of information and communication technology (ICT), why do we in most cases "download" from other experiences and think less of "uploading" to others our experiences? In my opinion, this is one of the major reasons for our failure as a nation seeking to be an economic giant with a strong industrial and technological base, and we all have a share of the blame. As academia, we are generally not business oriented toward showcasing our intellectual research products for economic rewards. More often than not, we discourage entrepreneurial initiatives by playing down on research efforts that

are directly applicable to solving problems and providing services, but which may not necessarily end up in publications. For instance, I know that we generally ignore commissioned research/service efforts in our considerations for promotion. I feel that this is counter-productive. Indeed, academia should be encouraged to engage in partnership initiatives with industries and other segments of the economy. We need to change the general attitude of Nigerian businessmen and manufacturers, who are not willing to invest in research for future economic gains. Rather, the orientation is generally towards immediate returns of investments, hence our manufacturing base is weak as we engage mainly in promoting importation of goods and ideas. Nigeria has for a long time promoted what some have referred to as a "Buying and Selling" economy. We must strive hard to reverse this inexcusable trend.

However, I must admit that the environment for bringing the academic and business communities together for the needed complementary roles has not been there for quite a while. This is where government has failed over the years. It is reasonable, therefore, to conclude that there has been a general cross border lukewarm attitude towards the transfer and harnessing of research output for social, economic and industrial gains. I know that someone will comment that government has demonstrated an initiative by establishing the Raw Materials Research and Development Council in the Ministry of Science and Technology. But the urge should go beyond a mere creation of an administrative unit. There must be a "window" deliberately created for effective consultations and realization of intended goals, and this must be through an improved innovative cooperation between laboratories and industries for coordinated corporate interactions and partnerships, and not bureaucracy. In addition, there must be a better funding of laboratories and adequate recognition and reward for excellence. In this regard, the question of protection and guarantee of reward for intellectual property, definition of

ownership of intellectual property, and coordination of agreements must be properly and promptly addressed. These requirements shall have to be attended to by all stake- holders namely, government, academia and private enterprises at all levels. I am aware of the existence of patent and copyright laws. But I am not sure if these laws guarantee adequate protection for intellectual property by the current applications. There is a general fear of "piracy/theft" of intellectual ideas and initiatives and these fears should not be wished off with the wave of the hand else the problem of transfer of research output for practical applications in solving our socio-economic and health problems will continue to remain with us. I can confirm that I have been a victim and so the worry is real. We must develop a strategy for improving our entrepreneurial skills and transfer of our intellectual innovations and ideas into what some have referred to as "Knowledge Economy". In one report on entrepreneurial development programme of the University of Wisconsin-Madison, USA, about 60 % of spin-off and start-up small-scale companies in the vicinity of the university employing about 7,000 people, derived their outlay from the Biological Sciences of the university.

I believe our university is a repository of intellectuals with great research and innovative ideas, as proven by our recent ranking as "First" in Research by NUC. But the concern here is about how much has been transferred or translated into Knowledge Economy. It is high time we took our university to a higher realm of superiority over others locally and internationally. I am aware that funding is an essential ingredient, but I believe we can also look inward and take some initiatives. In this regard, I am suggesting the creation of a unit in the university, with which researchers and inventors can interact for advice and possible transfer/translation of our research output and intellectual ideas and property into Knowledge Economy. If we do not have experts among ourselves, we should employ some from outside at least for a take-off. The

unit can also serve as an incubation platform for our products. This step should also reduce the fear of piracy/theft of intellectual property, standing as a dedicated and properly coordinated channel that can be monitored from within. A successful take-off should attract local and international grants in future. I am aware of the existence of the Linkages Unit of the university but I believe this assignment requires a professional touch and should, therefore, assume a separate identity for a definite function. This proposal is without prejudice for the expectation that there might be need for some collaboration in the long run. Our experience as mentioned earlier, of the loss of a glory that actually should have been that of this university and Nigeria should be informative in this regard.

LESSONS FROM RESEARCH EXPERIENCE

There is this phenomenon often referred to as "skill acquisition" or "learning on the job". In my experience, I have learnt to accept the need to be thorough and painstaking in the execution of research schedules, and be careful and cautious in drawing research conclusions. I have come to conclude that the quality of output may not necessarily correlate with the number of reports. In my opinion, a research output from a less thorough input, and often with inadequate and premature conclusions could lead to a chain of undesirable events, including misinformation and confusion. Permit me to illustrate with three incidents that occurred in my laboratory.

CASE 1: I refer you to my earlier report on the assessment of the application of the extract of the leaves of *Vernonia amygdalina* in brewing (M.Sc Projects). In one of our experiments, the student returned a result suggesting that the alcohol content was about 12 % (volume:volume). Ordinarily, the range of alcohol content of any brew using the brewers' yeast *Sacharomyces carlsbergensis* is 3.5-5 % for larger, and

may be up to 7.5 % for ale. A hasty conclusion here would suggest that we had broken into a new ground! But I insisted that the student should repeat the experiment, and which he did five times each time returning a value close to 12 %. I became worried and had to begin a thorough examination of the experimental set-up and assessment of the performance quality of the instruments used for taking measurements. Expectedly, the student became frustrated but he had no choice! In the end, our perseverance paid off as we found out that the source of the error was the alcohol used in setting the standard concentration curve. What was sold to us as absolute alcohol had been diluted, thus giving us exaggerated estimated values. What a relief for both the student and the supervisor!

CASE 2: This experience is amusing but also informative. One of my undergraduate students was to work on bacteria that required the use of blood agar medium. He was to collect blood from the abattoir for the preparation of the culture medium. On presentation of plates of his prepared medium they were full of growth of blood haemolysing bacteria. The medium was supposed to be sterile (free of microorganisms) so I queried his microbiological aseptic technique. Thrice he went back to the abattoir for fresh blood and each time he presented the same results claiming that he had followed all laboratory aseptic procedures. I then wondered if the Ile-Ife abattoir was not selling to us beef that did not meet public health standards, since the blood of a healthy cow was not supposed to contain microorganisms. Before raising any alarm, I decided to see things for myself. I followed the student to the abattoir to supervise the collection of blood sample, and also take some other notes that might help in unfolding the mystery! You guess what? The student had been collecting blood flowing on the slaughter slab rather than direct from the jugular cut. Again what a sigh of relief!

CASE 3: In this case, one of my M.Sc. students was working on some samples of soils collected for environmental studies. One of the plates presented with an unexpected overgrowth with organisms. I queried contamination resulting from poor aseptic technique. The student was asked to repeat the experiment each time submitting the same result. On a closer look, an observation was made of a gradual radial dispersion of discrete colonies of the organism from the point of inoculation and which excluded contamination and poor aseptic technique. I have since studied the organism to some more details and it would appear that a strain of bacteria with a new type of movement on agar plate has been discovered. Detailed analysis of the sequence of the DNA is being generated in collaboration with Professor Peter Golyshin of the National Research Centre for Biotechnology (GBF), Brauschweigh, Germany. About 80 % of the DNA sequence has been elaborated and reported to FASTA DNA sequence data bank (request ID No. 20050725073006-16087), using Smith-Waterman algorithm version 3.4t21 of May 14, 2003; (Smith and Waterman, 1981; Pearson and Lipman, 1988; Pearson, 1991).

You will agree with me that these examples that I have enumerated above constitute food for thought. The first two could have led to gross and damaging misinformation and confusion, while the third one could have led to a painful and probably irretrievable loss of useful discovery. In my opinion, thoroughness in research is imperative and should be encouraged. This can be achieved by balancing the game of number with that of quality and relevance. A well thought out and carefully executed research with one report/publication and a reasonably informative landmark should be more productive than a research with multiple reports and disjointed conclusions.

My humble suggestion, informed by my experience as stated above, is that it should be possible for us to evolve a system whereby an assessment of the quality of research output for reward through promotion would not be exclusively numerical i.e. based solely on the number of publications. I know that some universities abroad that have some formula for the translation of outreach services, particularly on commissioned research and transfer of innovative ideas, into publication credits. Apart from the reward by promotion, the move is bound to encourage the development of entrepreneurial activities to the advantage of the university in terms of fame and revenue generation capacity.

CONCLUSION

I have been on a sojourn into the world of microbes. Given the prevailing circumstances of my era, the achievements of my laboratory could appear hidden to be seen. But just like the microorganisms, you probably would need the microscope to elucidate full appraisal! Besides, I believe that a measure of achievement or success may not strictly speaking be seen on the pages of reports and publications, but resides in relevance and applications to solving problem. I feel fulfilled that I have made some modest contributions to knowledge and capacity development, but I regret that I have not been able to train a virologist. I keep my hopes open on this perhaps the opportunity may still come by!

Vice Chancellor, Sir distinguished audience, I have learnt a lot from the world of microscopic "giants", the first creation of life forms, the relics of the ages past, and the key of hope for the future. Have you too learnt from them? You better do, because there is greatness in smallness.

I thank you for listening.

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