UNIVERSITY OF PORT HARCOURT

MOLECULAR GENETICS: THE SOUL OF BIOTECHNOLOGY IN THE WAR AGAINST HUNGER, DISEASE AND POVERTY

An Inaugural Lecture

By

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DEDICATION

To my late mathematics teacher and father, Ezinna, Nze (Sir) Vincent Benson Oparanwata Osuji (KSJ), who laid the foundation for my education, saw me through to its pinnacle but narrowly missed this lecture. May his kind and humble soul rest in the bosom of the Lord, Jesus Christ, Amen.

ACKNOWLEDGEMENTS

I wish to express my profound gratitude to God and my parents, Ezinna Nze Sir Vincent Benson Oparanwata Osuji (KSJ; late) and Ezinne Lolo Lady Celestina C. Osuji, who sacrificed so much for my education. I wish to acknowledge the support I received from my wife, Genny and children, Akunna, Obieze and Kem. I also remember my daughter Udochukwu who has not had the right of her father's attention for reasons of man's inhumanity to man.

Let me express my gratitude to the Vice-Chancellor, Prof. Joseph Atubokiki Ajienka for granting me this opportunity to give my inaugural lecture at this time of my career. I am also grateful to Prof. Onyewuchi Akaranta, who continued to be a source of encouragement and support while I prepared for this academic exercise. I do appreciate the encouragement I received from the DVC Research and Development, Prof. Bene W Abbey and DVC Amin, Prof. E.C. Nduka.

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MOLECULAR GENETICS: THE SOUL OF BIOTECHNOLOGY IN THE FIGHT AGAINST HUNGER, DISEASE AND POVERTY

PROTOCOL

The Vice-Chancellor, Sir,

Members of the Governing Council here present,

Deputy Vice-Chancellors,

Registrar and other Principal Officers,

Provost, College of Health Sciences,

Dean, Graduate School,

Deans of Faculties,

My Fellow Professors and other Academic Colleagues,

Directors and Heads of Department,

Your Royal Highness,

My Lords, Spiritual and Temporal,

Great Students of Unique Uniport,

Friends of the University,

Distinguished Ladies and Gentlemen.

PREAMBLE

It would have been interesting to have this lecture on the topic "The more the look, the less is seen in Genetics". Unfortunately, the topic would not exhaust the capabilities of genetics to drive the world's very dynamic and powerful field of biotechnology. After careful review of the scope of my field, and its potential to turn things around, I decided to have my inaugural lecture topic reflect the scope and applications of my training. Hence the topic: **Molecular Genetics: The Soul of Biotechnology in the fight Against Hunger, Disease and Poverty**

In order to impart good grasp of this lecture, I wish to review few theories that would help us understand the workings of the subject matter. The topic of this lecture, though high-brow, in nature is targeted at helping us bring home the import of the new generation field, molecular genetics in driving biotechnology. Let us then zero our minds to make the best of this lecture.

ORIGIN OF MATTER AND ENERGY

Several theories have tried to explain how things started to be. The most popular of the theories explaining the beginning of all existence are the 'Big Bang' and the 'Creationist' theories. Other theories on this subject matter are not quite popular. According to the 'Big Bang' theory, the cosmic egg, which was smaller than the size of a hydrogen atom exploded to yield matter and energy. What remains unclear about this theory is how the 'Cosmic egg', being less than a hydrogen atom can hatch by its explosion all matter and energy. Subsequently, matter condensed to form atoms, elements, the universe and all that it contains, etc.

The 'Supernatural' theory, though seemingly less factual, appears to provide explanation to the unclear issues surrounding the 'Big Bang' theory. This theory is outside our current understanding of science since science is an attempt to measure, study and modify the natural world, The main thrust of this theory is that life came by the action of an eternal being, God. Within this definition are a broad range of beliefs. At one extreme are biblical literalists who believe that all life was created in its present form, including Adam and Eve as the first humans, as described in Genesis and with little or no evolutionary change since then (special creation). At another end are creationists who have no quarrel with evolution and believe it is God's method of creating life (theistic evolution), the view accepted today by several Christian denominations (Larson, 1997; Strahler, 1987).

Well, as for me, the 'Big Bang' and the creationist theories complement each other. Obviously each of them explains the other. Hence God may have said "Let there be..." and the 'Big Bang' launched something into being, the infinite point of the explosion being referred to by the evolutionist as

‘Cosmic egg’. The steps of creation outlined in Genesis, agree with the stages of the developments through the ‘Big Bang’ to the origin of life and its diversity as explained by Darwin (Robinson, 2002).

Origin of Life

The ‘Extra terrestrial theory explains that a meteorite carrying remains of organic matter and which impacted on the planet earth is the source of organic matter and by implication life in the planet earth. This is supported by the theory of ‘Organic Chemical Evolution’. Until the mid-1800's scientists thought organic chemicals (with a C-C skeleton) could only form by the actions of living things. A French scientist heated crystals of a mineral (inorganic chemical), and discovered that they formed urea (an organic chemical) when they cooled. Russian scientist and academic A.I. Oparin, in 1922, hypothesized that cellular life was preceded by a period of chemical evolution. These chemicals, he argued, must have arisen spontaneously under conditions existing billions of years ago (quite unlike current conditions).

Theories Contributing to Modern Biology

Modern biology and of course, biotechnology, are based on several great ideas, or theories such as: theory of thermodynamics, Homeostasis, Cell theory, gene theory and theories of evolution.

Thermodynamics covers the laws governing energy transfers, and thus the basis for life on earth. Two major laws are known: i) the conservation of matter and energy, and entropy; and ii) the universe is composed of two things: matter and energy.

Homeostasis is the maintenance of a dynamic range of conditions within which the organism can function. Physical factors such as Temperature, pH, and energy are major components of this concept.

The Cell Theory: Rudolf Virchow (in 1858): combined the ideas by Matthias Schleiden and Theodore Schwann that plant and animal tissues consist of cells and added that all cells come from pre-existing cells, and formulated the Cell Theory. The cell theory states that: (1) all organisms are made of cells; a cell is the structural and functional unit of organs, and therefore cells are organisms; and (2) cells are capable of self-reproduction and come only from preexisting cells.

The Gene Theory: James Watson (American scientist) and Francis Crick (British scientist) at Cambridge in 1953 developed the double helix model for deoxyribonucleic acid (DNA), a biochemical that had then been deduced to be the physical carrier of inheritance. Crick hypothesized the mechanism for DNA replication and further linked DNA to proteins, an idea since referred to as the **'Central dogma'** that anchors the gene theory. The theory has it that information from DNA "language" is converted into RNA (ribonucleic acid) "language" and then to the "language" of proteins. The central dogma explains the influence of heredity (which is coded in genes in DNA) on the organism (through proteins). The reality of this "central dogma" is very obvious in gene expression. Watson continued to make important theoretical contributions to genetics with a particular interest in development, until he turned his attention to neuroscience in the late 1970s. (Crick, 1988; Sherborn, 1995; Strathern, 1997).

EVOLUTION OF THE CELL AND LIVING ORGANISMS

Cell Evolution: Cellular life emerged on Earth in the form of primitive bacteria approximately 3.5 billion years ago. Bacteria are primitive organisms (each cell being 1-10 µm long) and organize their genes into a circular chromosome that lies exposed within the fluid environment (cytosol) of the cell. They are known as prokaryotes (from the Greek *pro*, meaning "before" and *karyon*, meaning "kernel" or "nucleus") because they contain a nucleoid region rather than a true nucleus where their genetic material is found. Bacteria diversified into various cell types within a billion years (Margulis, 1998). The different cell types had evolved numerous adaptive ways of extracting energy from the environment.

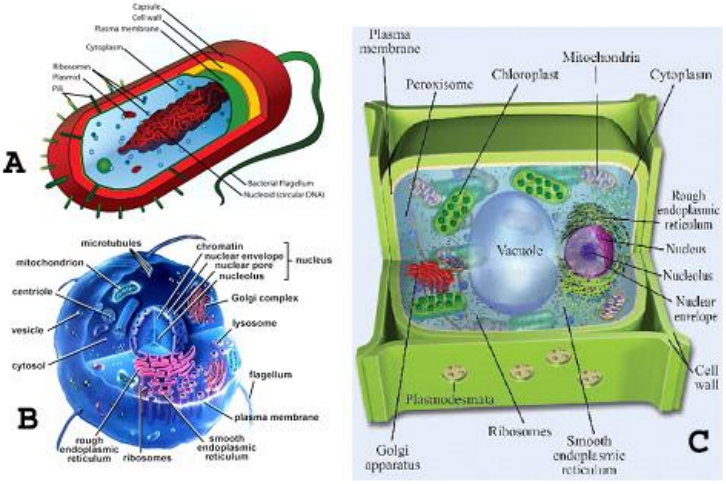


Fig 1: Typical schematic presentation of (A) prokaryotic and (B- C) eukaryotic cells; (B) Animal cell and (C) Plant cell

These various types included: first the anaerobic/fermenting archaebacteria, secondly the oxygen-producing photosynthetic cyanobacteria, and thirdly aerobic/respiring bacteria which is able to utilize the new oxygen-rich atmosphere. In addition some bacteria had become flagellated.

About two billion years ago, advanced cellular organisms or "eukaryotes" (from the Greek *eu*, meaning "true" and *karyon*, meaning "kernel" or "nucleus"), whose deoxyribonucleic acid (DNA) is sequestered within a separate membrane-bound nucleus emerged (Fig. 1). They are commonly larger than prokaryotic cells and measure about 10-100 µm long. These cells also contain an extensive internal membrane system, a cytoskeleton, and different kinds of membrane-bound organelles, including mitochondria (the "power factories") and, in algae and plants, plastids (sites of photosynthesis). All multicellular life, including plants, animals, most large algae and fungi, are composed of eukaryotic cells; some microbes, such as yeasts, unicellular algae and protozoa, are also eukaryotes (Mader, 1998). See Fig. 2 and Table 1 for more details.

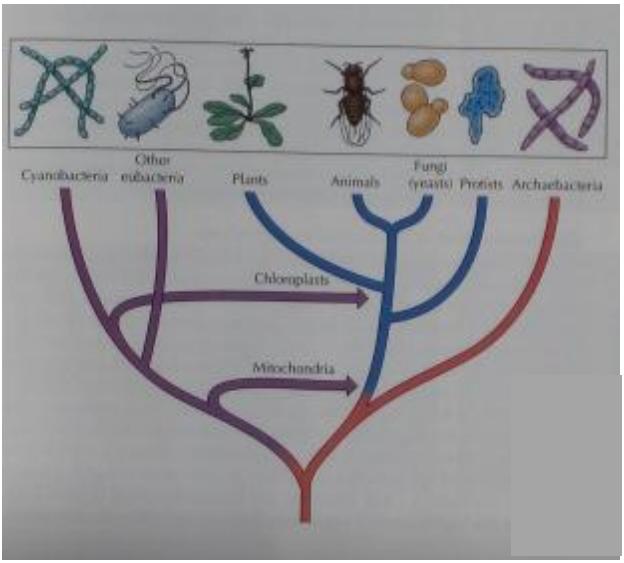


Fig 2: Lines of evolution of Present day cells from a common prokaryotic ancestor, which gave rise to archaebacteria, eubacteria and eukaryotes. Mitochondria and chloroplasts originated from the endosymbiotic association of aerobic bacteria and cyanobacteria with the ancestors of eukaryotes. Robinson, 2002 with permission).

Table 1: Organelles and structures found in Cells

Structure	Function
Nucleus*	Contains genetic material
Ribosomes	Protein synthesis
Endoplasmic reticulum	Synthesis/modification and transport of proteins and lipids
Golgi apparatus	Processing, distribution of proteins, lipids
Lysosomes	Digestion of substances in cell
Peroxisomes	Digestion and detoxification
Mitochondria*	Energy generation
Chloroplasts*	Photosynthesis
Flagella/Cilia	Cell movement
Vacuole and vesicle	Storage of cellular substances
Centriole	Cytoskeletal organization
Plasmids*	Reproduction, disease resistance, etc.

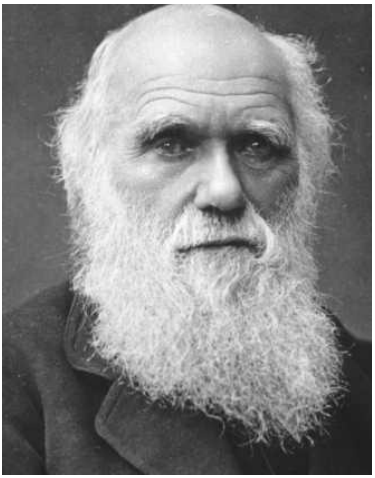
* Organelles/structures that contain genes in DNA structures (Adapted from Robinson, 2002)

Multicellularity: Multicellularity depicts where two or more cells together constitute an organism. In most multicellular organisms, there are many different types of cells that perform specialized functions. In animals, for instance, bone cells are for support, glandular cells secrete hormones, epithelial cells are for protection, stem cells are for growth and production of other cells, etc. while in plants the three basic types of cells are parenchyma, collenchyma and schlerenchyma. These cells make up tissues which make up organs (protective, package, transport, meristematic, photosynthetic, storage,) which compose systems and then organisms. However, for molecular

cytogenetic studies, meristematic tissues are of utmost interest but thin-walled living tissues (parenchymatous tissues) are also very useful depending on the type of information required.

DIVERSITY OF LIVING THINGS

***Darwinian Theory of Evolution*:** Darwin is generally recognized as the single greatest thinker in the history of biology, whose contributions provided the basis for understanding the immense diversity that characterizes the natural world (Browne, 1996; Lewis *et al;* 2002; Robinson, 2002). Darwin and his contemporaries, especially Alfred Russel Wallace, showed how one organism gave origin to another, from one primitive level of existence to a less primitive level until life got to the pro-advanced stages.. The Darwinian Theory explains to a great extent the present day origin of biodiversity and gives insight into the role of molecular genetics in genetic engineering and biotechnology.



Charles Darwin, *English naturalist, 1809–1882*

Darwin was born February 12, 1809, into a wealthy English family. His lifelong interest in natural history led him to embark, at age twenty-two, on a five-year voyage to South America aboard the HMS *Beagle* as the ship's naturalist. Darwin collected a wealth of specimens and made observations of both the living species and fossils he encountered. Darwin was particularly struck by similarities he observed between the species found on the Galapagos Islands off the western coast of South America, and species of the mainland. He also noted differences and similarities among species found on the numerous islands of the Galapagos. The evidence suggested each species had not been independently formed by the Creator, but rather had diverged from a smaller group of common ancestors. The diversity he observed was obviously caused by **hybridization, phenotypic plasticity and genetic polymorphism.**

Darwin pondered these ideas in conjunction with two ideologies. The first was geologist Charles Lyell's theory of uniformatarianism. This mechanism suggested that Earth was much older than previously believed, a fact which Darwin saw as providing the requisite time for the steady accumulation of change that would turn one species into another. The second ideology was from *An Essay on the Principle of Population,* in which economist Thomas Malthus contrasted the potential for exponential increase in human population with the much slower increase in food supply. Malthus suggested that competition, disease, war, and famine kept the human population in check. Darwin saw that this principle provided the selective force needed to bring about change in a species. (Hartl and Jones, 1998; Robinson, 2002)

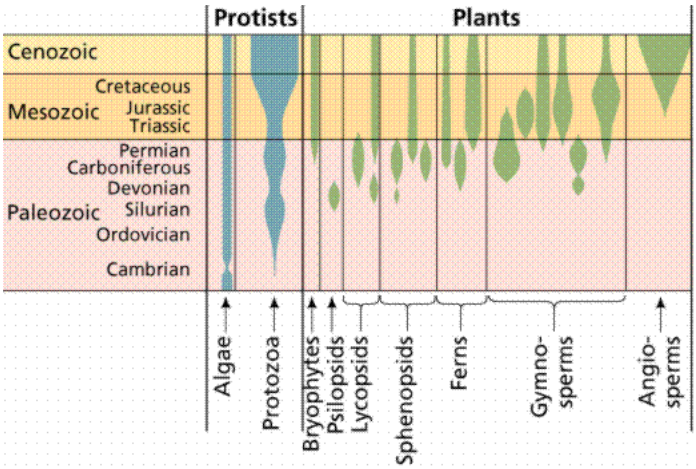


Fig. 4: The fossil records of some protist and plant groups. The width of the shaded space is an indicator of the number of species. Image from Purves *et al*., Life: The Science of Biology, 4th Edition, by Sinauer Associates (www.sinauer.com) and WH Freeman (www.whfreeman.com), used with permission

Evolution by Natural Selection: Darwin developed his ideas into the principle of natural selection (between 1837 and 1838). This principle combines the phenomena of struggle, heritable variation, and differential reproduction to explain natural selection through survival. He proposed that in all species, limited resources lead to a struggle for existence, either against other members of the species, or against the environment. Naturally, members of a species vary from one another. Some of those variations influence the success of an organism's struggle. Organisms with more useful variations leave more offspring (biodiversity), who inherit those variations and therefore are more capable of coping with environmental stress (Robinson, 2002).

As this process continues over time, with the attendant successive rounds of struggle, variation, and differential reproduction, the population will become increasingly well adapted to the environment. Those that cannot adapt die. This leaves behind those that can cope, which are therefore seen as having been selected by nature, hence the term 'Natural selection'.

Vice Chancellor Sir, my personal opinion in this matter is that evolution is simply the process that unfolds creation. In other words, God said "Let there be..." and the process known as 'evolution' ensues. Based on my researches and the works of other scientists, I have the conviction that edible *Musa* i.e. plantain, *Musa paradisiaca* L. and banana, *M. sapientum* L. are triploid evolutionary products of *M. acuminata* Colla and *M. balbisiana* Colla (Charlesworth, *et al*., 1994; Dover, 1982; Heslop-Harrison, 2000; Heslop-Harrison and Schwarzacher, 2007; Lavergne *et al*., 2010; Meagher, *et al*., 2005; Osuji *et al*., 1996a, 1996b, 1997a, 1997b, 1997c, 1997d; Osuji, 1998; Osuji *et al*., 1998a; Teuber and Zorn, 1982).

When the process of taxonomic divergence occurs naturally, it is called evolution but when it is caused by man's deliberate effort to create diversity, it is called genetic engineering. Taxonomic groups evolve various characteristics, otherwise they would be placed in just the same group. Let us then examine what constitutes a character.

CHARACTERS AND CHARACTERISTICS OF LIFE

A character can be explained as a feature or attribute that is stably-expressed under normal conditions. Every life, be it unicellular or multicellular, micro organism, animal or plant has a set of characteristics, which serve as the basis for its life and identity, i.e. an organism is an embodiment of its characters. Observable characteristics of life include:

i) irritability, ii) movement, iii) feeding, iv) growth, v) respiration, vi) reproduction and vii) adaptation. All these characteristics must be present for the cell or organism to be sustainably alive. In order to understand the organism well enough to domesticate or manipulate it, these characteristics must be properly understood.

The characteristics could be quantitative (if they can be measured or counted) or qualitative (if they cannot be measured or counted). Quantitative variables can be statistically processed whereas qualitative characters can only be estimated by scoring to bring them to a quantitative platform before they can be properly analyzed. Some examples of quantitative characters are: yield, height, number of seeds, size of seed/fruit/leaf, height of plant, weight, girth etc. while qualitative characters are: taste, medicinal property, disease resistance, drought tolerance, some biochemical and environmental phenomena; distribution, pesticide bioaccumulation (Nwachukwu and Osuji, 2008) etc.

Whereas some of the characteristics are physically observable (such as yield, height, length, thickness, colour, shape, texture, ornamentation, number, distribution (Nyananyo and Osuji, 2007; Onyeachusim *et al*., 2006; Osuji, 2006; Osuji *et al*., 1996b, 1997a,1998b), some are only assessed with optical aids (Anyanwu and Osuji, 2001, 2002; Okoli and Osuji, 2008; Osuji and Nsaka, 2009; Osuji and Ndukwu, 2005; Osuji *et al*., 1996a, 1996b, 1997a, 2009; Osuji and Agogbua, 2010). Some other characters are not optically observable but can be evaluated by other means (e.g. Nwachukwu and Osuji, 2007; Osuji and Eke, 2005; Osuji and Nwachukwu, 2006). Yet, another group of characters is time-related (Osuji *et al*., 2006a, 2006b; Ekanem and Osuji, 2006; Osuji and Owei, 2010). Despite the variability of characters (Figs. 5 and 6), the field

that offers the means of studying their transmission from one generation to another is Genetics.

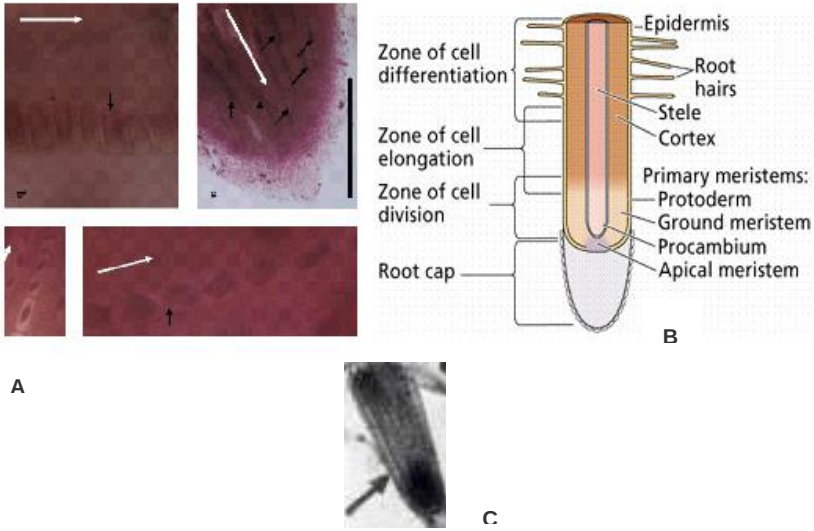


Fig. 5: A) The root tip and component tissues of yam, Dioscorea rotundata (Culled from Osuji and Agogbua, 2010), (B) Typical Root tip and (C) Raphide bundle containing calcium oxalate crystals.

Fig: 6 Bananas showing clear evidence of morphological diversity

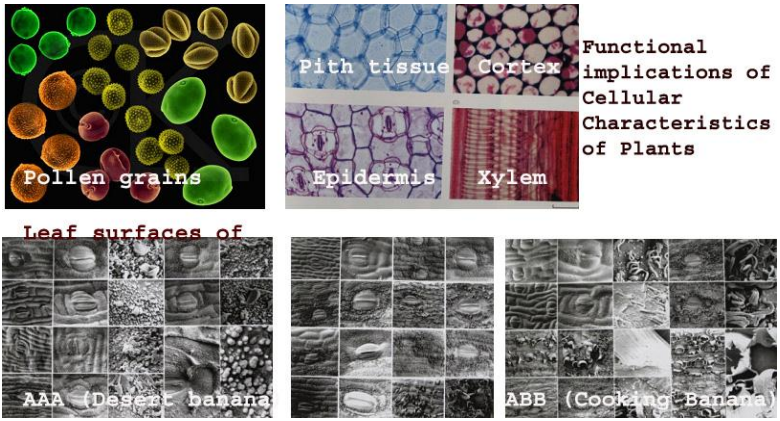


Fig. 7: Functional implications of cellular characteristics of plants (Osuji and Heslop-Harrison, unpublished)

GENETICS

This is the science of heredity which began with observation of the features/characteristics/traits of living things. It seeks a precise explanation of the characteristics of biological organisms, mechanisms of inheritance and the expression of inheritance. In other words, it endeavours to define the various characteristics, unravel the basis and processes of transmission of the characters from parents to their progeny (be it a person, an animal, a plant or microbe).

Before Gregor Mendel, two of many misconceptions blurred peoples' understanding of heredity. The first was that one parent contributes most to an offspring's inherited features. Aristotle contended it was the male by way of a fully formed homunculus, inside a male sperm or pollen grain in plants. The second was the concept of blended inheritance, the idea that parental traits become mixed and forever changed in the offspring. He just became interested in the work he did

monastery (at Brno) and accessed Darwin's "*On the Origin of Species*".

Mendel's work comprised his observations while cultivating pea (beans) for his monastery. This apparent beginning of genetic studies resulted in the development of the early principles of inheritance. Mendel worked on crossing of single lines of characters/traits (i.e. monohybrid crosses), double and multiple lines) dihybrid and multihybrid, test and back crosses of pea.

Mendel showed that a character was controlled by a factor, which was later termed 'gene'. He explained that a gene has two alleles and there are two forms of an allele: the dominant and the recessive forms. The dominant form expresses itself in the presence of another dominant form or a recessive form. On the other hand, the recessive form can only be expressed in the absence of a dominant form (allele) of the gene. Mendel established the law of segregation, which showed that during gamete formation, the two alleles separate or segregate and enter different gametes. As a result, each pollen grain, ovule, sperm or ovum carries only one of each pair of parental alleles. Cross-pollination and fertilization between pure-breeding parents with opposite traits result in F_1 hybrid zygotes with two different alleles, one from each type of parent, for each type of trait. Different alleles of a gene segregate each from the other and enter into different gametes.

The law of independent assortment shows that in a dihybrid or multihybrid cross, each pair of alleles segregates independently so that in the gametes, one member of each pair is equally likely to appear with either of the two alleles of the other pair or pairs of alleles (of other genes). In other words, when alleles of two or more genes are involved in gamete formation, each pair of alleles assorts independent of any other

pair. The pattern of separation of members of one pair into gametes is independent of the pattern of separation of another pair. Mendelism is anchored on physically expressed characters.

Deviations from Mendelism

There are often several complexities in relating genotype (the genetic composition) to phenotype (physical appearance or expression). These make difficult, the application of Mendelian principles in certain circumstances. Some examples are as follows:

i) ***Incomplete dominance:*** If a hybrid is identical to one of its parents for a trait under consideration, the allele that was expressed is deemed dominant and the one that was not expressed is deemed recessive. Where the hybrid is not identical to any of the two parents for a trait under consideration the situation is referred to as incomplete dominance. For instance, a cross between pure early blooming and pure late blooming pea yielded a hybrid that was neither early nor late blooming.

ii) ***Co-dominance*****:** This is where the hybrid displays the features or traits of both parents. Here, alternative traits of both parents are visible in the hybrid. An example is in humans where some of the complex membrane-anchored molecules that distinguish different types of red blood cells show co-dominance.

iii) ***Multiple Alleles*****:** A gene may have more than two alleles, e.g. The ABO blood types are determined by three different alleles.

iv) ***One gene may have multiple visible expressions***: Mutation can affect genes. When mutation affects a gene in a way that the gene's phenotype changes, the result may be more than one wild-type characteristic. For this reason, a gene with only one wild-type allele is said to be 'monomorphic' while a gene with more than one wild-type allele is polymorphic. An example is the mouse Agouti gene.

v) ***Recessive Lethality*:** This is a situation where a gene's expression leads to death. For example, people who are homozygous for the recessive S allele often develop heart failure due to stress on the circulatory system.

vi) ***Pleiotropy*:** This is where a gene affects or controls more than one visible trait. For instance, the S allele of the β-globin gene affects more than one trait. The hemoglobin molecules in the red blood cells of homozygous SS individuals behave aberrantly after releasing their oxygen. i) instead of remaining soluble in the cytoplasm, they aggregate to form long fibers that deform the red blood cell from a normal biconcave disk to a sickle shape, which obstructs the flow of blood by clogging small vessels; ii) the sickled cells are very fragile and easily broken; iii) SS homozygotes are resistant to malaria because the sickled cells when infected by *Plasmodium falciparum* break down before the organism has a chance to multiply.

vii) ***Epistasis*:** This is where two or more genes can interact to determine one trait such as a petal colour, seed coat colour, a chicken's feathers, or a dog's fur. In these cases,

(where homozygosity of a recessive allele of one gene is required to mask or hide the effect of another gene). Another example is ocular-cutaneous albinism (OCA) in which two albino parents could produce non-albino children. This demonstrates that homozygousity for a recessive allele of either of the two genes can cause OCA.

viii) ***Modifier genes*:** This is where, for instance, sometimes a genotype is not expressed at all. In other words, even if the genotype is present, the expected phenotype does not appear. Some other times, the trait caused by a genotype is expressed to varying degrees or in a variety of ways in different individuals (in a manner that could be caused by environment or chance). This same situation arises when expression of a gene depends on its penetrance or expressivity.

ix) ***Cytoplasmic / maternally inherited genes*:** This involves genes which are not located in the nucleus. The two organelles in the cytoplasm that have DNA are plastids (e.g. chloroplasts) and mitochondria (Fig. 8). Their genes are inherited from the female gamete because the female usually contributes its cytoplasm in sexual reproductions (Careel *et al*., 2002; Faure *et al*., 1994).

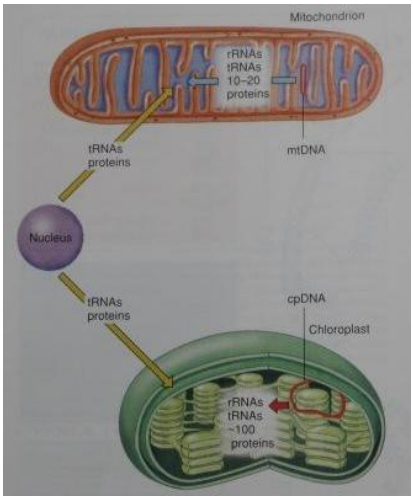


Fig. 8: Cellular domains of heritable genes. Although the nucleus contains the major set of genes, the chloroplast and mitochondria contribute genes that are cytoplasmically inherited.

CYTOGENETICS AND THE TYPICAL CELL

The Cell is made up of protoplast covered by the cell wall (in plants and prokaryotes) and cell membrane (in animals). The cell wall encloses the protoplast. Excluding the vacuoles, the living component of the protoplast is called protoplasm, which comprises a fluid matrix called cytoplasm into which are immersed the double membrane nucleus and organelles.

The organelles found in cells are mitochondria, chloroplast (in plant cells), endoplasmic reticulum, golgi apparatus, ribosomes, lysosomes, peroxisome and centrioles (in animal cells). The nucleus contains most of the genetic material (DNA) in the cell. However, mitochondria and chloroplasts are known to possess DNA also. The DNA located in the chloroplast and mitochondria possess genes,

which contain coded information for production of major proteins and enzymes required in them (Fig. 9). For now our major attention is to be given to the nucleus.

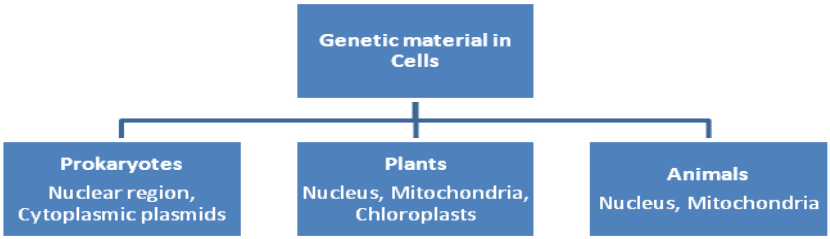


Fig. 9: Distribution of genetic materials in cells

The nucleus is the largest structure in the cell. It is usually located nearly centrally in young cells and animal cells but close to one side of the cell in old differentiated plant cells. The nucleus is surrounded by a double membrane structure called nuclear membrane, which adjoins the endoplasmic reticulum. The space inside the nucleus is filled by a fluid matrix called nucleoplasm in which are embedded chromatin strands called chromosomes (coloured '*chroma'* bodies '*soma'*), and nucleoli (singular: nucleolus). Prokaryotic cells contain only one circular chromosome located in a region of the cell called the genophore or nuclear region, which lacks nuclear membrane. Conversely, the nucleus in eukaryotic cells contain linear (sets of) chromosomes, the total number of which range from four to several tens in various species. Chromosomes bear, in their DNA, factors of heredity called genes. How is this possible?

The Chromosome Theory of Inheritance

The clearest insight into the organization of the chromosome was first derived from close observation of the chromosomes of prokaryotes (bacteria) and later confirmed with more studies of the eukaryotic cells. Information revealed by various studies (including partial digestion of chromatin with micrococcal nuclease and electron microscopy) show that a set of histone proteins namely: histone 1 (H1), histone 2A (H2A), histone 2B (H2B), histone 3 (H3) and histone 4 (H4) are involved at the most remote level of organization of the chromosome. Two each of histones 2A, 2B, 3 and 4 come together in an octameric bundle and are wrapped around by naked DNA strand to form a structure or kernel that consists of 146 base pairs of DNA wrapped 1.65 turns around the histone octamer, called nucleosome core particle. The DNA turns around the nucleosome core particle is locked or sealed by histone 1, which holds it from loosening (Kornberg, 1974). The complex of nucleosome and histone 1 constitutes the chromatosome (comprising 166 base pairs long DNA and is 10 nm in diameter; Cooper, 2000). See Figs. 10 and 11.

A short strand of DNA called linker DNA connects two adjoining chromatosomes (also regarded as the basic structural unit of chromatin). The entire structure from one end of the long DNA strand to the other end appears like beaded string. The organization of the chromatosomes constitutes the initial compaction of the DNA.

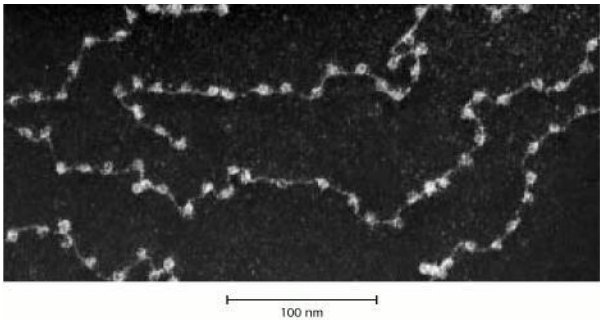


Fig. 10: Chromatin Structure. An electron micrograph of chromatin showing its "beads on a string" (i.e nucleosomes attached to linker DNA) character. [Courtesy of Dr. Ada Olins and Dr. Donald Olins.]

Special non-histone proteins called scaffold proteins attach to the linker DNA portions of the beaded structure to form chromatin. There are more than one thousand types of non-histone proteins, which are involved in a range of activities including DNA replication and gene expression. Further condensation of the DNA is through chromatin coiling into thin (30 nm) fibre, whose thickness may increase depending on 1) extent of coiling and 2) stage of the cell cycle. Thus it is the chromatin that further compacts through coiling and folding to make up the chromosome. Therefore, the chromosome is composed of a pair of DNA strands (molecules) and associated proteins.

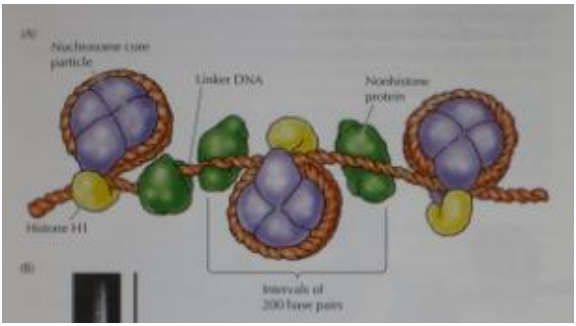


Fig. 11: The appearance of chromatin showing nucleosome core particles and linker DNA in association with scaffold proteins.

There are two types of chromatin namely euchromatin and heterochromatin. Euchromatin is relatively decondensed (mostly about 30 nm thick fibres) and make up about 90 % while heterochromatin is very condensed and make up about 10 % of total chromatin. In nondividing (i.e. interphase) cells, most of the euchromatin are distributed throughout the nucleus. During this 'resting' (non-mitotic) period of the cell cycle, genes are transcribed and the DNA is replicated in readiness for cell division. The euchromatin is largely organized into loops containing approximately 50-100 kb (kilobase i.e. x1000 bases length) of DNA. About 10 % of the euchromatin is more decondensed and contains the genes that are being actively transcribed. Contrastingly, the heterochromatin is very condensed and consequently relatively thicker resembling chromatin of dividing (or mitotic) cells.

Chromosome Features

The major features of chromosomes are their number, structure and behavior (Okoli and Osuji, 2008). The structure is derived from the condensation or compaction of chromatin during cell

division and localization of the primary constriction in relation to the length of the arms of the chromosome (Fig. 12:). Thus, when the primary constriction is at the centre of a chromosome, it is said to be metacentric. Depending on the distance of the constriction from the centre i.e. based on the relative length of the arms of the chromosome, a chromosome could be submetacentric, subacrocentric, acrocentric or telocentric. Some chromosomes associated with nucleoli organizers have another constriction called the secondary constriction that adjoins a chromatin appendage called satellite. Individual chromosomes can be distinguished or identified by karyotype analysis (Fig. 13:)

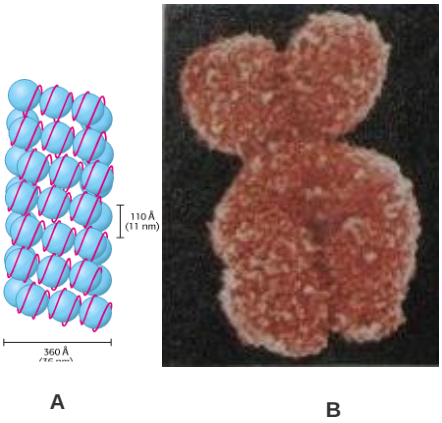


Fig. 12: Higher-Order Chromatin Structure. A) A proposed model for chromatin arranged in a helical array consisting of six nucleosomes per turn of helix. The DNA double helix (shown in red) is wound around each histone octamer (shown in blue). [After J.T. Finch and A. Klug. *Proc. Natl. Acad. Sci. USA* 73(1976):1900.]; and B) SEM image of a metaphase chromosome.

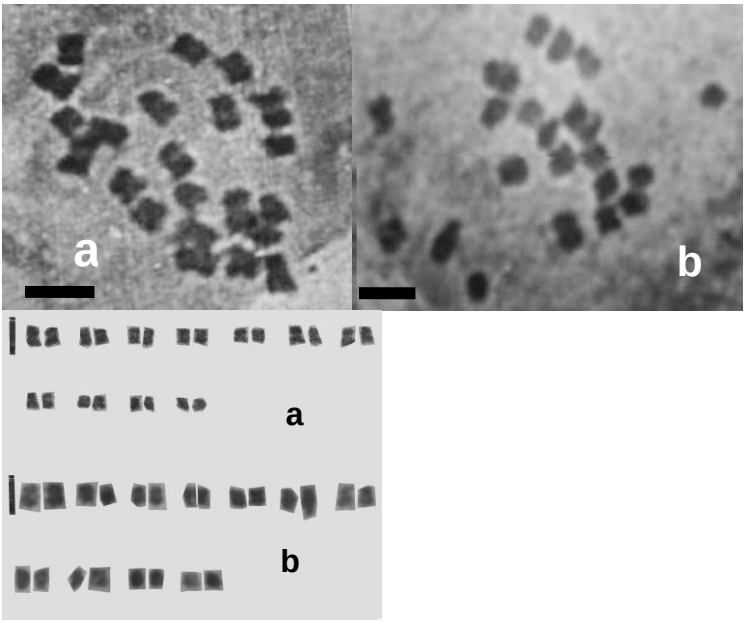
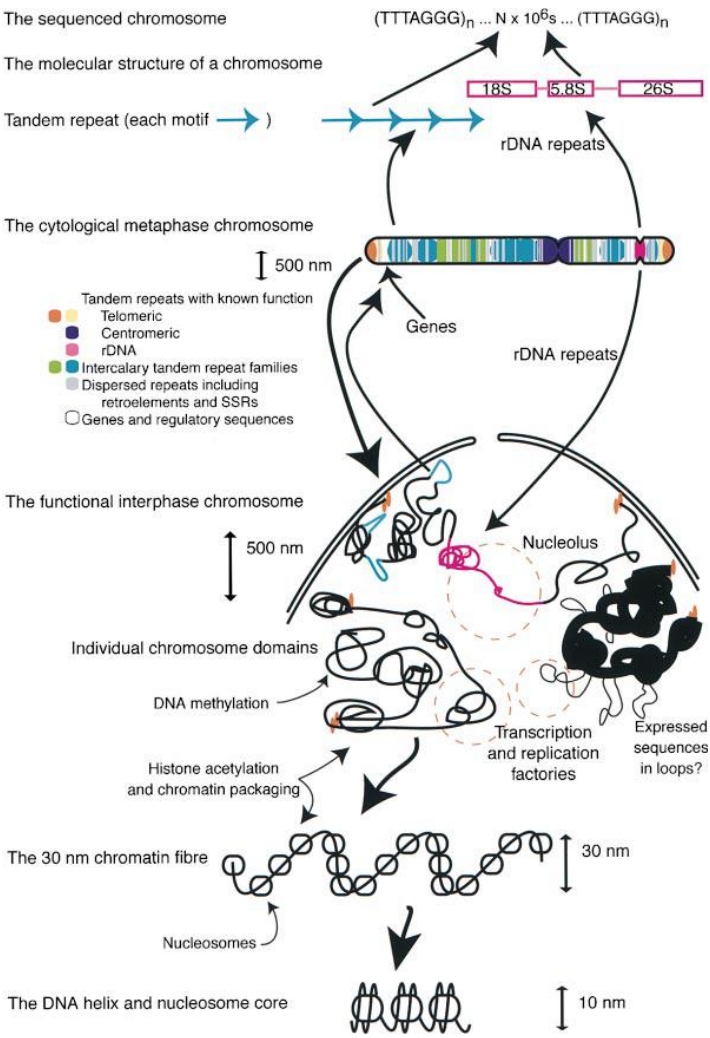


Fig. 13: Karyotypes of a) *Musa acuminata* Colla and b) *M. balbisiana* Colla. Scale bar = 5 µm. (Osuji *et al*., 2006).

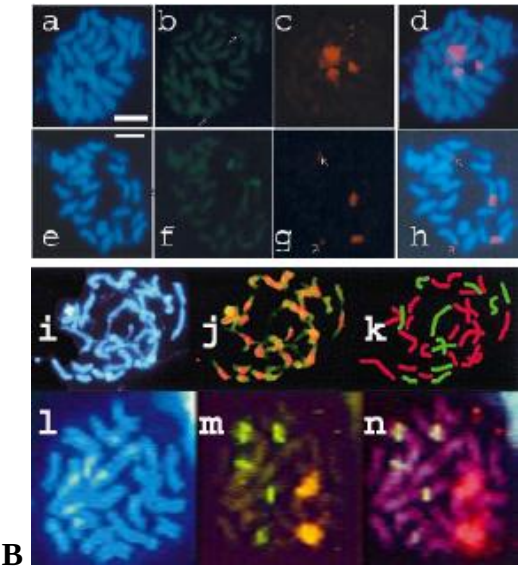
MOLECULAR CYTOGENETICS HELPS TO MAP DNA TO SPECIFIC GENOMIC DOMAINS

Fluorescent *in situ* hybridization of labeled DNA samples to chromosomes obtained from root tips of plants is used to track genes and chromosomes (Fig. 14). It could be applied to samples from blood, tissue biopsies, buccal scraps, amniotic fluid, cultured cells, etc. and extensively used to diagnose various forms of human health conditions. The same DNA samples can also be used by application of Southern blotting methods to provide information relating to genetics, phylogeny and taxonomy. The DNA sample is generally digested using a restriction endonuclease and then subjected to electrophoresis in a horizontal agarose gel.



A

Fig. 14: A) A Model illustrating the workings of molecular genetics on Plant Chromosomes. Culled from Heslop-Harrison (2000) with permission;



B

B) *In situ* hybridization of 5S and 18S-5.8S-25S rDNA sequences on mitotic chromosomes of two cultivars of *Cucumeropsis mannii*; a) DAPI image, b-c) filtered images and d) sites of rDNA on mitotic chromosomes of the cultivar with oblong-round fruit; e) DAPI image, f-g) filtered images and h) sites of rDNA on mitotic chromosomes of the cultivar with cylindrical fruit. Osuji *et al*., 2006; i-k) *in situ* genomic hybridization of genomes A and B DNA to plantain chromosomes (Osuji *et al*., 1997d); and l-m) *in situ* hybridization of rRNA genes to chromosomes of banana hybrid (Osuji *et al.*, 1998a).

Every species, contains a specific number of chromosomes in the nucleus of each of its cells. The number of chromosomes in a germ cell or gamete of an organism is usually half of the number in its body (or somatic) cell. Hence gametes are normally haploid. There are special cases where a

gamete may have two sets of parental chromosomes (i.e. dihaploid state). The number of chromosomes in each cell is constant for every normal cell of the organism.

The chromosome complement for a species is commonly in two sets, each being inherited from either of the parents. Depending on the number of sets of chromosomes derived from parents, a cell or organism can be diploid (one set from each parent), triploid (one set from one parent and two sets from the other parent), tetraploid (four sets made up of two sets from each parent) etc. Most funji and algae are haploid (i.e. only one set of chromosomes), hence they reproduce mostly vegetatively or sexually through dikaryotization (especially in fungi). Other ploidy levels are pentaploid, hexaploid, heptaploid, octaploid, nonaploid, decaploid, etc. However, it is rare to have a species that has up to six sets of chromosomes as in the bread wheat (*Triticum aestivum*). The representation of the chromosomes of a species is its karyotype. Despite the number of chromosomes, the sum of the DNA in the karyotype or nucleus reflects the genome.

Sets of chromosomes have peculiar ways of behaving during cell division based on their structural make-up. Hence, during gamete formation, pairs, triplets, quadruplets etc. of homologous chromosomes hybridize (i.e. come together harmoniously). Whereas most form rods, others could present themselves as circles, crosses, etc. The pairing behavior is unique for each species. On the other hand, during the second half of cell division, chromatids representing daughter chromosomes separate to the two opposite poles appearing in various shapes. The pairing and separation behaviour of chromosomal complements of a species is constant. The only source of change or alteration of chromosome behavior of a species is chromosomal aberration.

The Genome

This is the totality of the genetic information carried by a cell of an organism. It reflects the complete set of chromosomal DNA and includes the total number of genes of an organism (see Figure 15 below). Study of the genome is called genomics. It yields information on genetic (structural, functional and organizational) variation of organisms down to the molecular level. Genome analysis yields important information such as: comparison of organisms at the molecular level, and for detecting the evolutionary relationships of genes, gene families and species. It is applied in the tracking of effects of mutations and as well in the counteraction and reversion of genetic events through genetic engineering (Müller, 2008).

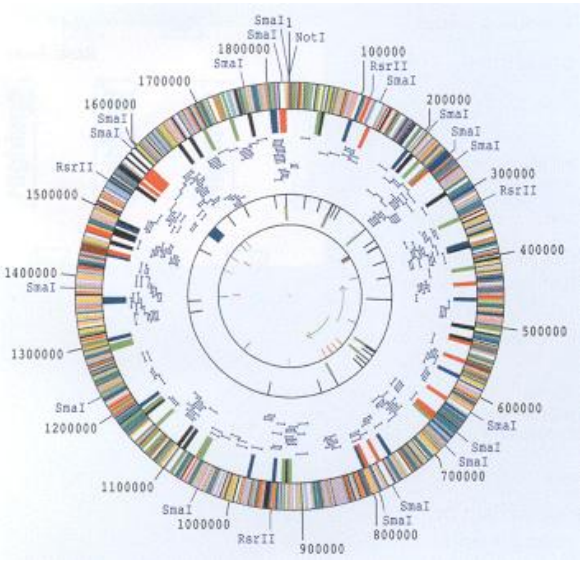


Fig. 15: Diagramatic representation of a Complete Genome. The diagram depicts the genome of *Haemophilus influenzae*, the first complete genome of a free-living organism to be

sequenced. The genome encodes more than 1700 proteins and 70 RNA molecules.

The Gene

However, DNA, whether in chromosomes, chloroplast or mitochondria is composed of functional polynucleotide sequences (i.e. segments or units) called genes as well as non-functional sequences, which may be pseudo-genes, non-coding DNA sequences (introns) or intergenic spacer sequences (Bennett and Leitch, 2004; Buhariwalla *et al.,* 2005). The genic or functional DNA sequences are of utmost interest in molecular cytogenetics because they contain the functional sequences, which are expressible as characteristics of the organism.

In other words, a gene is a segment of the DNA molecule, which itself is a polymer of repeating (monomeric) groups known as nucleotides. This therefore implies that a gene is a sequence of nucleotides, which is part of the larger DNA molecule. Whereas this explanation shows a gene as a unit of structure, genes are known to control phenotypic features and genotypic functions. Therefore a gene can also be defined as a unit of phenotypic expression, a unit of function or a unit of physiological activity.

Gene Expression processes: Flow of Genetic Information from Nucleic acids to Protein

In order to have a clear impression of the process of gene expression, it is proper to understand the nature and relevance of DNA, RNA and protein in the 'central dogma'. These biochemicals are the media for information storage and expression for life.

The DNA Molecule

One DNA molecule runs through the length of each chromosome (Heslop-Harrison, 2000) or a plasmid. The DNA molecule has two right handed double helical (double stranded) structures and is tightly buttoned at a position referred to as the primary constriction. The DNA molecule in the chromatin (as earlier mentioned) is the structure that contains the genes.

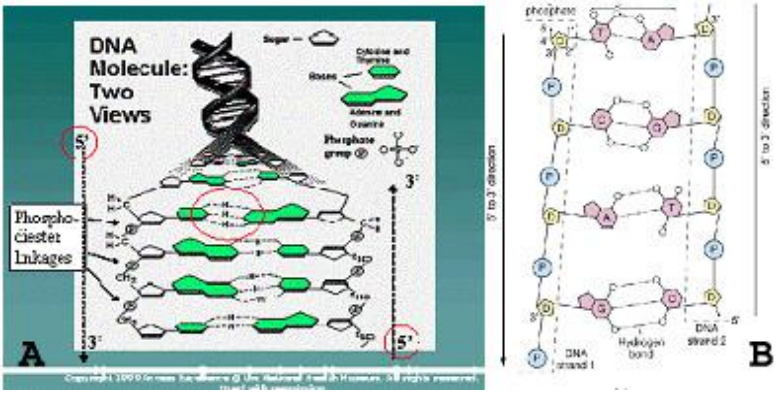


Fig. 16: Double helical structure of a segment of DNA molecule: A) DNA Structural details showing the derivation of the double helix from the phosphodiester-linked polynuleotide chains; and B) the molecular structure of a segment of DNA molecule.

In-as-much-as a DNA molecule could be several millions of nucleotides long; only four repeating units of nucleotide compose it (Fig. 16). A nucleotide is a compound that is composed of 5-carbon sugar (deoxy-ribose), phosphate group and nitrogen base residues. They are named after their bases. The bases are: adenine, thymine, cytosine and guanine.

Information in DNA can be reproduced through semi-conservative or conservative replication

The RNA Molecule

Another type of nucleic acid known as ribonucleic acid (RNA) exists but is not a component of chromatin. RNA molecule is single stranded but also composed of four basic nucleotide residues. Unlike the DNA, the nucleotides found in RNA molecules have ribose sugar (instead of deoxy-ribose in DNA), a phosphate group and one each of four nitrogen bases. The nitrogen bases in RNA are similar to those of DNA except that thymine (in DNA) is replaced by uracil (in RNA).

There are three different types of RNA namely: messenger RNA (mRNA), transfer RNA (tRNA), and ribosomal RNA (rRNA). Whereas they are mostly relevant in the cytoplasmic domain of the cell, templates of their nucleotide sequences are found in the chromosomal DNA (Osuji *et al.*, 1998).

Transcription of DNA into RNA

The differences between DNA and RNA are simple. In RNA, thymidylic acid (T) is replaced by uracylic acid (U). The difference between 'T' and 'U' is that 'T' has a methyl group attached to its nitrogen base while 'U' has just hydrogen residue attached in the same position in place of methyl group. A fragment of the strand of DNA molecule serves as a template for assemblage of mRNA which is a notable agent of gene expression.

For this to happen, a segment of a DNA strand serves as a template for assemblage of messenger RNA (mRNA). RNA polymerase (enzyme) positions itself at the promoter region of the gene sequence to be copied and sequentially goes

through the transcription unit (i.e. reading frame) of the sequence. The transcription unit or gene's reading frame is transcribed into RNA following the pairing complementarities of the nucleotides. However, where ever there ought to be a T, a U is loaded on the RNA molecule being assembled. Once the polymerase reaches the termination unit of the DNA segment, it slides off the DNA strand and the process is terminated. The RNA is then transferred to the cytoplasm for translation. This may be after post transcription processing. See Fig. 17 below.

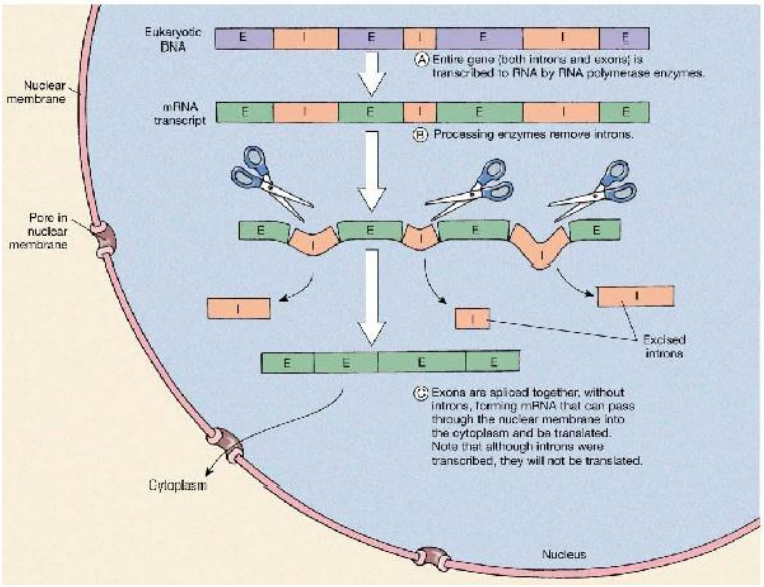


Fig. 17: Transcription from DNA to RNA and post transcriptional processing of mRNA in plants. Post transcriptional processing of mRNA shows that the mature RNA moleculae is shorter than the primary RNA transcript.

In eukaryotes, RNA post transcription processing produces a mature messenger RNA from the primary (pre-) mRNA molecule. RNA processing is necessary because, the DNA template sometimes contains interposing or intervening non-coding DNA sequences (introns) in their reading frames which split the coding sequences (or exons) into two or more fragments. The pre- mRNA transcript has to be processed into one single mature frame that contains no introns.

RNA processing in prokaryotes adopts simple mechanisms whereas there are different forms of RNA processing in eukaryotes as follows: i) processing by Splicing mechanisms, ii) Alternative splicing, iii) RNA editing and iv) RNA degradation. In processing by splicing mechanism, RNA processing modifies only the 5' (front end) or 3' (back end) of the primary transcript, leaving untouched the information contained in the rest of the mRNA.

Alternative splicing occurs frequently in genes of complex eukaryotes and provides an important mechanism for tissue-specific and developmental regulation of gene expression. Typical examples are genes that encode transcriptional regulatory proteins. Since several pre-mRNAs contain multiple introns, different mRNAs can be produced from the same gene by different combinations of 5' and 3' splice sites. In other words, different mRNAs can be assembled by combining different exon units transcribed from the same DNA reading frame.

RNA editing refers to processing procedures, other than splicing, that alter the protein coding sequences of some mRNAs. This unexpected form of RNA processing was first discovered in mitochondrial mRNAs of tripanosomes, in which U residues are added and deleted at multiple sites along the mRNA molecule. This process of RNA editing has been

recently described in mitochondrial mRNAs of several organisms, chloroplast mRNAs of higher plants, and nuclear mRNAs of some mammalian genes (Cooper, 2000).

The result of processing mechanism, alternative processing and RNA editing is mature messenger RNAs, which then direct protein synthesis. The final step in the processing of RNA is the eventual degradation of mRNA within the cell after protein synthesis. The factor that regulates intracellular level of mRNA is the balance between synthesis of protein and degradation of RNA molecules. The degradation of most eukaryotic mRNA is initiated by shortening of the poly A tails. This is followed by removal of the 5' cap and degradation of the mRNA by nucleases acting from both ends.

Translation of information in the mRNA into Protein

In the cytoplasm, the mRNA is moved to the ribosome, which serves as the site of protein synthesis (otherwise called translation). The three types of RNA (i.e. mRNA, tRNA and rRNA) play distinct roles. Proteins are synthesized from mRNA templates by a process that has been carefully conserved throughout the ages. The process has been part and parcel of the evolutionary process.

During translation, all mRNA templates are read from the 5' to the 3' direction, and polypeptide chains are assembled from the amino tip to the carboxyl terminus. The determination of amino acid residue's position in the polypeptide chain is specified in the widely accepted genetic code. Translation is executed at the rRNA location with tRNA serving as adaptors between the mRNA template and the amino acid residues, which are being polymerized into protein through polypeptide bonding (Fig. 18).

There are twenty types of amino acids that form proteins. These twenty types of amino acids are joined in several repeating sequences to compose the several thousands of proteins found in cells of bacteria, plants and animals. However, different tRNAs share similar overall structures though each type loads one particular amino acid.

The central importance of protein synthesis in cell metabolism is reflected in the fact that cells contain numerous ribosomes; eg. *E. coli* cell contains 20,000 ribosomes (25 % dry weight of the cell) whereas actively growing mammalian cell contains about 10 million ribosomes. Prokaryotic ribosomes have 16S rRNA and 21 proteins comprising the small ribosome subunit, the large 50S subunit being made up of 23S and 5S rRNAs and 34 proteins. The subunits of eukaryotic ribosomes are larger. The small (40S) subunit of eukaryotic ribosomes is composed of 18S rRNA and about 30 proteins while the large (60S) subunit is composed of 28S, 5.8S and 5S rRNAs (Osuji *et al*., 1998a) and about 45 proteins.

Translation is divided into three stages namely: initiation, elongation and termination. The ribosome has three sites for tRNA binding namely P (peptidyl), A (aminoacyl) and E (exit) sites. The initiator methionyl tRNA is bound at the P site. The first step in the initiation process is the binding of a specific initiator methionyl tRNA and the mRNA to the small ribosomal subunit. The large ribosomal subunit then joins the complex to form a functional ribosome (or polysome) on which the elongation of the polypeptide chain proceeds. After the initiation complex has formed, translation proceeds by elongation of the polypeptide chain. The elongation mechanism is similar in both prokaryotes and eukaryotes and proceeds until a stop codon (UAA, UAG or UGA) is

translocated into the A site of the ribosome. Cells do not have tRNAs with anticodons complementary to these termination signals. What cells have are release factors that recognize the signals and terminate protein synthesis.

Termination of synthesis is followed by dissociation of both the tRNA and mRNA from the ribosome. Messenger RNAs can be translated simultaneously by several ribosomes. Once one ribosome has moved away from the initiator site, another can bind to the mRNA and begin synthesis of a new copy of the same protein.

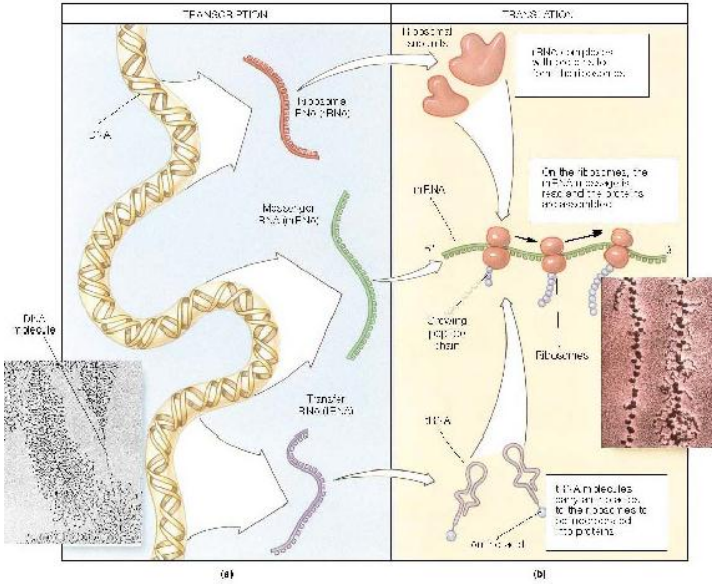


Fig. 18: The process of gene expression showing stages from transcription of information in DNA to RNA and translation of the same information to protein which is mobilized for cellular or extracellular function.

The Genetic Code

The DNA complement of a genome has all the information required for the fully functional life of the organism. The totality of the information is contained in the linear arrangement of the four types of nucleotide in DNA or the DNA dialect (i.e. A, G, C and T) and RNA or RNA dialect (i.e. A, G, C and U). In proteins, the same set of information is represented by the linear orderly combination of twenty amino acids. In this code, a triplet codon represents one amino acid. It is important to note that Watson and Crick produced the now accepted list of twenty amino acids that are genetically encoded by DNA or RNA sequence over lunch one day at a local pub (Hartwell, *et al*., 2000). Amino acids that are present in only a small number of proteins or polypeptides or in only certain tissues or organisms did not qualify as standard building blocks of proteins and hence are excluded from protein forming amino acids.

Several thousands of proteins exist but are composed of only twenty amino acid residues. Experiments have shown that groups of three nucleotides represent the twenty amino acids. Each nucleotide triplet is called a codon. Three triplets (UAA, UAG and UGA) denote the stop or nonsense commands/codons. The stop codons do not encode any amino acid thus they terminate translation. Biochemical manipulations revealed codons for individual amino acids (Fig. 19).

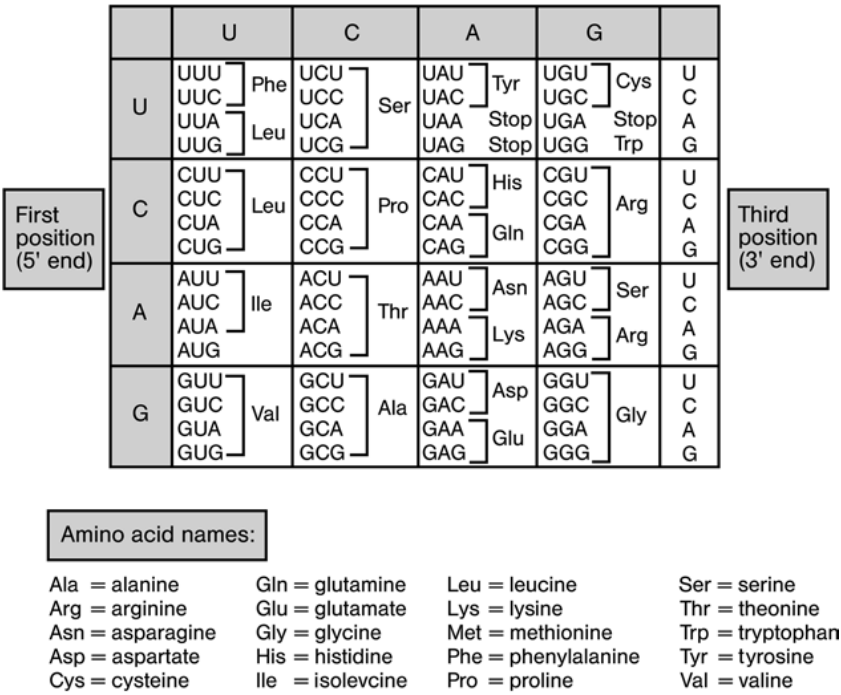


Fig. 19: Details of the genetic code and the amino acids represented by the nucleotide triplets. (Culled from Müler, 2008).

Since the code comes into play during protein synthesis (i.e. during the decoding of the messenger RNA to polypeptide), the code is usually presented in the RNA dialect. The genetic code is degenerate, i.e. more than one triplet combination often represents one amino acid. If the sequence of nucleotides in a gene or its mRNA transcript is known, the sequence of amino acids in the corresponding polypeptide would be easily deciphered. Mapping studies have confirmed that a gene's nucleotide sequence is collinear with its

corresponding peptide's amino acid sequence. Genetic analyses have revealed that non-overlapping codons are set in a reading frame. Consequently, each nucleotide is part of only one codon. The designated starting point for each gene establishes the reading frame for the triplets.

In reading the transcript of a gene, the machinery of the cell scans through the mRNA from a fixed starting point that establishes the reading frame. In certain contexts, the triplet AUG which specifies the amino acid methionine serves as the initiation or start codon. As such it often marks the spot in the nucleotide sequence of a mRNA where the code for a particular protein begins.

It is important to note that mutations can modify the message or information encoded in a sequence of nucleotides in three ways namely: frameshift, missence and nonsense mutations. Frameshift is when nucleotide insertions or deletions alter genetic instructions by changing or altering the reading frame. Missense mutation involves the change of a codon for one amino acid to that of a different amino acid (e.g. a change of GUU to GGU changes codon for valine to codon for glycine). This kind of mutation gave rise to the sickle cell condition. Nonsense mutation is one that changes the codon for an amino acid to a stop codon (e.g. a change of UAC to UAA changes codon for tyrosine to stop codon). These and other mutations affect genetic information and often lead to evolutionary changes and modified organisms.

However, mutation can be used as a means of producing modified genes that yield new or modified proteins (or end products) that may be of interest. Hence, molecular cytogenetics uses mutation as a means of breeding modified organisms. Having understood molecular genetics, we need

now to know about biotechnology in order to understand how they can work together.

BIOTECHNOLOGY

The word biotechnology, coined in 1919 by Karl Ereky, applies to the interaction of biology with human technology (Nill, 2002). For some thinkers, biotechnology is the means or way of manipulating life forms (i.e. organisms) to provide desirable products for man's use. According to Biotechnology Industry Organization (2007), biotechnology started with use of biological processes and *technology* to solve problems or make useful products. Basic biotechnology such as growing of crops and production of animals to provide a stable supply of food and clothing began 10,000 years ago. Biological processes of microorganisms to make useful food products, such as pap, have been ongoing for more than 6,000 years. It involves such simple practices as bee keeping, cattle rearing, making of pap, oil bean for salad, all locally fermented products, beverages, bread, cheese, to preserve dairy products and as well, such high precision practises as bionanotechnology, recombinant DNA technology (or genetic engineering), (Nill, 2002),etc.

However, the *Chambers Science and Technology Dictionary* defines biotechnology as 'the use of organisms or their components in industrial or commercial processes, which can be aided by the techniques of genetic manipulation in developing e.g. novel plants for agriculture or industry (Evans and Furlong, 2003)'. From all these efforts to define biotechnology, biotechnology ought to be broadly defined as 'the development and utilization of biological processes, forms and systems for obtaining maximum benefits to man and other forms of life; i.e. the science of applied biological processes.

More recently, biotechnology can be more appropriately described as the use of cellular and biomolecular

processes together with appropriate technologies to solve problems or make useful products. It includes application of scientific and engineering principles to the processing of materials by biological agents to provide goods and services (OECD, 1989; Dubey, 2004). It is multifaceted and could be described as where science and technology meet. It has an expanded scope, dimension, direction, application, input and more importantly output, which covers but are not limited to areas listed in the Table 2 below.

Table 2: Some areas of biotechnology

Area of Interest	**Products**
Recombinant DNA technology (Genetic Engineering)	Fine chemicals, enzymes, vaccines, growth & other hormones, antibiotics, interferon, Genetically Modified Organisms, etc
Biomass / Waste treatment & Utilization	Single cell protein, mycoprotein, alcohol and biofuels
Plant and animal cell culture	Fine chemicals (alkaloids, essential oils, dyes, steroids, etc.), Somatic Embryos, encapsulated seeds, interferon, monoclonal antibodies,
Nitrogen fixation	Biofertilizers or microbial inoculants
Bioenergy / biofuels	Hydrogen (via photolysis), alcohols (from biomass), methane (biogas) Produced from wastes and aquatic weeds), etc.
Biocatalysts	Fine chemicals, food processing, biosensor, chemotherapy
Fermentation	Acids, enzymes, alcohols, antibiotics, fine chemicals, vitamins, toxins (i.e. biopesticides, etc.
Process engineering	Effluent, water recycling, product extraction, novel reactor, product harvesting

Global Application of Biotechnology

Biotechnology is practiced at different levels. The simplest involves manipulation of the environment of living organisms to make them deliver certain products. This is widely practiced in breweries and the wine industry where certain materials of containers are used for maturation of wines to yield peculiar tastes, aromas and flavours. Another simple application of biotechnology involves induction of changes, usually by encouraging or inducing mutation in organisms used in the fermentation process in making beer, wine, confectionaries, etc. Mutants offer variation of the expected output in such industries and are therefore of great value in introducing variety in industrial end products.

In recent times, revolution in biotechnology has occurred due to several new complementary innovations. For instance, techniques have been developed to produce rare and medicinally valuable molecules, which are used to: (i) change hereditary traits of plants and animals; (ii) diagnose diseases; (iii) produce useful chemicals; (iv) clean up and (v) restore polluted or contaminated environment etc. For these reasons, biotechnology has great impacts on: Food and Agriculture, Industry, Environment and Health.

***i) Food and Agriculture*:** Enhancement of agricultural crop and animal resource transportation, production and conservation.
***ii) Industry*:** Production of industrial products with particular qualities that drive the market.
***iii) Environment*:** Pollutant cleaning (bioremediation) using genetically modified organisms with enhanced capability to extract chemicals from the environment.

***iv) Health*:** Synthesis of medically active biochemicals, which are applied for treatment purposes (e.g. gene therapy) and to control medical conditions.

v) Aesthetics/Recreation: Organisms of specific features are also produced for specific recreational interest. Some chimeras, like the blotched mouse shown, are created through genetic modification techniques like gene targeting. Some of these Genetically modified organisms can be ordeered through the internet (Figs. 20 and 24).

Fig. 20: GloFish, the first genetically modified animal to be sold as a pet (Eenennaam, 2008).

Owing to rapid development of procedures, processes and applications, there seems, at present, to be no difference for example, between pharmaceutical firms and biotechnological industry. Approved products and renewed public confidence make it one of the most promising areas of economic growth in the future. Today there are genetically engineered crops, animals, humans (ethically sensitive) and medical and industrial products.

Cells and Biological Molecules as the foundational structure for biotechnology

We know that cells are the basic building blocks of all living things. The simplest living organisms such as most prokaryotes

(e.g. bacteria and blue-green algae) and smaller eukaryotes (e.g. yeast), each consist of a single, self-sufficient cell. However, some of them comprise multicellular organizations. Complex creatures such as plants, animals and humans, are made of many different cell types, each of which performs a very specific task in the organism. ***In spite of the extraordinary diversity of cell types in living organisms, what is most striking is their remarkable genetic similarity. This unity of life at the cellular level provides the foundation for biotechnology.***

All cells have the same basic design, are made of the same construction materials and operate using essentially the same processes. Deoxyribonucleic acid, the genetic material of most living organisms, directs cell construction and operation, while proteins do all the work. Because DNA contains the information for making proteins, it directs cell processes by determining which proteins are produced and when. The means of achieving this is translation as earlier treated.

All cells speak the same genetic language as shown in the RNA dialect of the genetic code. The DNA information manual of one cell can be read and implemented by cells of other living organisms. Since a genetic instruction to make a certain protein is understood by many different types of cells, technologies based on cells and biological molecules give us great flexibility in using nature's diversity to achieve desired ends.

In addition, cells and biological molecules are extraordinarily specific in their interactions. Consequently, products of biotechnology can often solve specific problems, generate gentler or fewer side effects and have fewer unintended/unwanted consequences (against the fears of opponents of Genetically Modified Oganisms 'GMOs'). The terms 'Specific', 'Precise' and 'Predictable' best describe recent-time biotechnology output.

Sequencing and Sequence Maps of Polynucleotides (DNA and RNA) and Polypeptide (proteins) molecules lead the way

A sequence refers to pictorial representation of the order of amino acids in a protein molecule, nucleotides in a DNA molecule, and monosaccharide or oligosaccharide components in a glycoprotein/carbohydrate molecule. The term 'sequencing' as applied to polynucleotides is the process used to obtain the sequential arrangement of nucleotides in the DNA backbone (Baurens *et al*., 1996). The cleavage into fragments of DNA molecule is followed by separation of those fragments, which can then be sequenced individually by one of the following methods: (i) a chemical cleavage method followed by Polyacrylamide Gel Electrophoresis, 'PAGE' or capillary electrophoresis, (ii) a method consisting of controlled interruption of enzymatic replication methods followed by PAGE, (iii) a dideoxyl method utilizing fluorescent "tag" atoms attached to the DNA fragments, followed by use of spectrophotometry to identify the respective DNA fragments by their differing "tags" (which fluoresce at different wavelengths). This (fluorescent tag) variant of the dideoxyl method can be automated to "decipher" large DNA molecules (and genomes). Such automated machines are sometimes called "gene machines." Sequencing of DNA was first done in the mid-1970s by Frederick Sanger (Nill, 2002). Currently Polymerase Chain Reaction 'PCR' machines are popularly used for this purpose (Fig. 21).

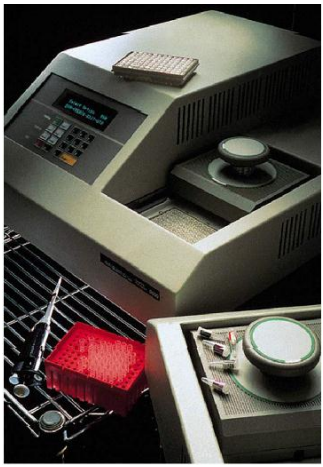


Fig. 21: Polymerase Chain Reaction 'PCR' Machine is used in sequencing DNA.

Fabrication, Carpentry and Joinery as applied to DNA works in Molecular Genetics

The discovery of two particular enzymes has revolutionized application of molecular genetics in biotechnology. This development is manifest in the field of recombinant DNA technology or genetic engineering. The two enzymes are i) Restriction endonucleases and ii) Ligase.

i) Restriction endonucleases, a class of enzymes that cleave (cut) DNA at a specific and unique internal site or location along its length. These enzymes are naturally produced by bacteria that use them as a defense mechanism against viral infection. The enzymes chop up the viral nucleic acids and hence their destruction. There are several types of restriction endonucleases each of which cuts DNA at a particular location. The location of cut by the restriction enzyme is controlled by the nucleotide sequence of the DNA target molecule. Each restriction enzyme has a particular nucleotide sequence or combination at which it cuts. Therefore whereas one may cut

where there is e.g. GAATTC (see Figure 22 below), another may cut where there is GGCGCC, etc. For this reason each of them is restricted to cutting DNA at the location of a particular nucleotide sequence.

Restriction endonucleases are important tools in genetic engineering, enabling the biotechnologist to cut open a DNA molecule in order to splice new genes into the cut location(s) (where a restriction endonuclease has created a gap via cleavage of the DNA).

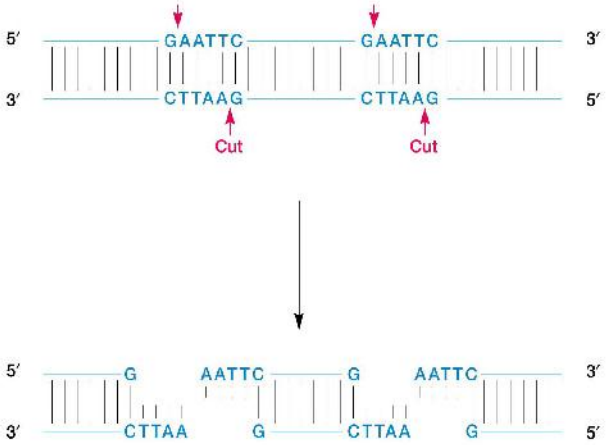


Fig. 22: Restriction endonuclease cuts DNA strands at positions of specific polynucleotide sequences. Each enzyme has a specific polynucleotide sequence through which it cuts and is different from sites where other enzymes cut.

ii) Ligase, An enzyme used to catalyze the joining of single-stranded DNA segments. Ligase causes ligation, which is the formation of a phosphodiester bond to link two adjacent bases separated by a nick in one strand of a double helix of DNA (deoxyribonucleic acid). The term can also be applied to blunt-end ligation and to the joining of RNA strands.

Cloning

The term originates from the fact that cells which are genetically and biologically homogeneous, usually produced vegetatively or asexually from one original cell consist of a clone of cells. Thus cloning represents the production of many copies of an engineered DNA, usually in a genetically and biologically homogeneous group of cells derived asexually from one initial cell. The amplification of a specific cloned gene or genes, coupled with a marked increase in production of their protein products, makes it relatively easy to extract and purify these proteins in the laboratory. In a typical cloning procedure, a suitable plasmid (vector) is selected in which to insert a desired gene (donor DNA). Both donor DNA and vector are digested with the same restriction enzyme, and then incubated together with ligase to join the donor DNA fragments with the plasmid. The result is a recombinant plasmid that contains the desired DNA fragment. For DNA to be incorporated into a bacterium, the bacterial cells would first be made competent by treatment with calcium. With this treatment, the bacterial cells take in the DNA by phagocytosis. The recombinant plasmid is then used to transform a host bacterial cell, creating a new genetic strain of the bacteria that stably maintains the recombinant plasmid.

Transformation/genetic engineering of cells and organisms in Recombinant DNA technology

This involves incorporation of an exogenous DNA into another organism. The DNA may be a polynucleotide sequence representing a gene from the cell of an organism different from the cell into which it is to be incorporated. For example, the gene for disease resistance or human insulin gene may be obtained from the cellular domain of a source organism and incorporated into a cell or cells of another organism. The

exogenous DNA is subsequently induced to integrate into the bacterial plasmid DNA. As a component of the plasmid DNA, the gene expresses itself in the bacterial cell. If the gene is to be incorporated into a plant or animal cell, the process would follow one of the following procedures: i) electroporation, ii) use of gene gun, iii) micro-injection or iv) agro-bacterium infection, etc.

i) Transformation by Electroporation: Electroporation, also called electroporesis or electropermeabilization is a process utilized to introduce a foreign gene into the genome of an organism. In this method, a suspension of exponentially growing host cells is mixed with a solution of recombinant DNA molecules and exposed to a high electric field for a few milliseconds. The brief high voltage direct-current (dc) electrical pulse alters the structure of the cell membrane and temporarily causes formation of tiny holes (called "micropores") in the surface of the cells or protoplasts. After the gene enters the cell via the temporarily created micropores, the electrical pulse ceases, and the micropores re-seal so that the gene(s) cannot escape from the cell. The cell then incorporates (some of) the new genetic material (genes) into its genome. Once the exogenous gene is in place inside the host genome, it expresses itself by creating whatever product (i.e. a protein) the exogeneous gene codes for. In 1995, the U.S. company Dekalb Genetics Corporation received a patent for producing genetically engineered corn via introduction of a foreign gene into corn cells via electroporation.

ii) Use of Particle bombardment Gun or Gene gun: This technique is alternatively known as i) particle bombardment, ii) biolistic process, iii) particle gun process, iv) microprojectile bombardment, v) gene gun or vi) particle acceleration. A gene gun is an equipment that is used to send

micro-projectiles such as tungsten pellets coated with DNA fragments (i.e. genes) into plant cells or tissue at very high speed. Once the equipment is shot, the pellets with the genes are launched through the cell membrane or outer surface of the cell structure into the cell. Once inside the cell, the DNA unbinds from the pellets and incorporates into the genome of the host cell.

This technique is most suitable for those plants which hardly regenerate and do not show sufficient response to gene transfer through *Agrobacterium* spp. This method is also useful in delivering exogeneous DNA into pollen grains. The pollen grains being so transformed are then used for directed or controlled pollination. The result is genetically engineered seeds, which can then be planted and nursed into grown plants. Cells that are transformed via this means are regenerated in tissue culture.

iii) Micro-injection: Transgenes or exogenous DNA, which can drive a cellular or subcellular function can be introduced into a cell by injection. The process is called micro-injection because it involves use of a glass micropipette to deliver the DNA into a cell. The tip of glass pipette used for micro-injection measures about 0.5 mm and therefore resembles an injection needle. The types of cell that can be micro-injected are relatively large cells, such as: oocyte, ovum or egg and embryos of animals. Such cells are relatively larger than most other cells and can be seen even with little or no visual aid. Once done, the foreign DNA is induced to integrate into the nuclear genome of the injected cell.

The ovum can be implanted into the real or foster female animal for sexual fertilization. It could also be artificially inseminated before implantation into a real or foster mother for development of the zygote into full-fledged pregnancy. Several sites of insertion of the foreign gene may

result. Some of the copies of the foreign gene could be degraded by enzymes in the cell. Once successful, all of the cells of the zygote and resultant embryo contain the transgene often as multiple inserts. As the embryo develops into an offspring and later in subsequent reproductive generations in which the transgenic offspring participates as a parent, stabilization of the transgenesis takes place and results in true lines of the transgenic animal.

iv) Agrobacterium tumefasciens *and* Agrobacterium rhizogenes *Plasmids-mediated transformation*: *Agrobacterium tumefasciens* is a bacterial species that is ordinarily free-living. It exists around the top soil and constitutes part of the bacterial flora within 15 cm level of the base of plants, especially dicot plants. This bacteria has a tumour-inducing (or Ti) plasmid that is capable of being transferred by the bacterium into plant cells by conjugation. When a transfer of the plasmid into a plant cell takes place, the plasmid induces the plant cell to grow into a tumour-like structure called crown gall. The genes in the plasmid, once in the plant cell, would commandeer the cell to produce special proteins [opine, octopine and nopaline] in large quantities. The *Agrobacterium tumefasciens* in turn feeds on the proteins produced by the swollen plant cell.

Based on the known ability of *Agrobacterium tumefasciens and Agrobacterium rhizogenes* to transfer the tumour-inducing plasmid into plant cells, such plasmid has been reconstructed or engineered and used to deliver choice (or selected) genes into *in vitro* cultured plant cells. In the process of construction of the plasmid, undesirable portions of the plasmid DNA is excised while DNA fragments representing genes of interest are incorporated into the plasmid, which is introduced into the plant cells of interest by infection with the genetically engineered bacteria. This means has been highly

exploited in biotechnological production of several drugs composed of secondary metabolites and alkaloids, e.g. atropine (*Atropa belladonna*), visnagin (*Ammi majus*), artemisnic acid and arteannin B (*Artemisia annua*), cinchonine and cinchonidine (*Cinchona ledgeriana*), etc.

v) Liposome-mediated gene transfer: Liposomes are cytosolic organelles or particles also called liquid bags. Many plasmids are enclosed in them. By using polyethylene glycol (PEG), they may be stimulated to fuse with protoplast in several plants like carrot, tobacco, petunia, etc. Due to endocylosis of liposomes, the lipid bilayer membrane fuses on contact with cell membrane and the DNA enclosed in them is liberated into the protoplast. This technique has advantages which include i) low toxicity, ii) long stable storage of DNA fragments in liposome, iii) protection of DNA or RNA fragments from nucleases, iv) high level of reproducibility and v) applicability in various types of cells.

vi) Direct Transformation: In order to transform mammalian cells, it is necessary to precipitate the DNA with calcium phosphate and mix with the cells to be transformed (Fig. 23). The DNA fragment passes through the cell membrane and integrates randomly with the chromosomes. Using this technique, a selective marker can be linked up with the DNA fragment to be cloned before introduction into mammalian cells. The transformed cells can then be separated from their cell line after plating them on selective medium.

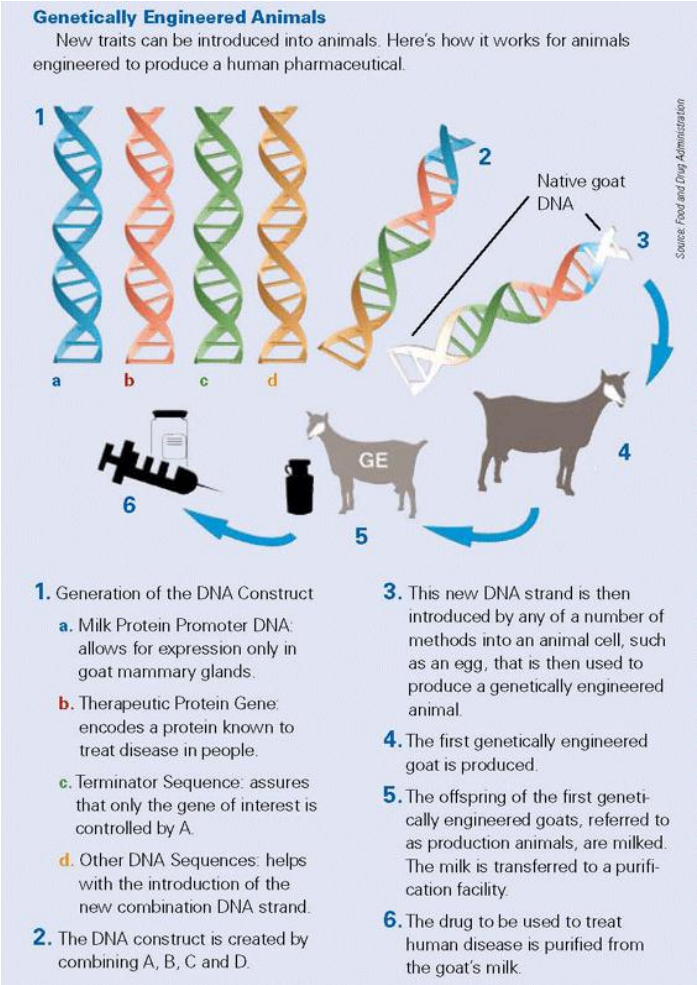


Fig. 23: Direct transformation of cow with DNA fragments representing various genes of interest.

***vii) Site-Directed Mutagenesis*:** There are techniques used to mutate a specific portion of the genome in order to get novel results or products of enormous value. Such site-specific

mutations could be used to silence a gene, or even to alter single amino acids in a protein in order to improve its properties. For such modifications, point mutations i.e. alteration of single nucleotide, is done on a specific portion of the gene. It can also be done by knock-out mutation in which an oligonucleotide corresponding to mutated or unwanted gene and its adjacent regions (about 15-20 nucleotides long) is used to replace it. See Fig. 24.

Fig. 24: Genetically modified mice. A) Blotched chimera mice, B) transgenic mice for sale through the internet and C) knock-out mice produced through knock-out mutation. All were produced for their aesthetic values.

viii) *Protoplast fusion:* This involves the fusion of protoplast of two cells. If the cells are similar, this creates a polyploid of the species being so treated. If the protoplasts belong to two different species, the treatment serves as a means of creating new superior organisms. The product of protoplast fusion is called a cybrid because it is a hybrid produced by combining the genetic and cytoplasmic components of two parent cells. Cybrid technology is used to combine good properties of species one with nuclear gene superiority and the other with superior cytoplasmic properties.

Artificial Propagation

Once a cell has been genetically modified through any of the described means of transgenization (or transformation), it has to be sustained or supported. Cells of most eukaryotes normally exist in multicellular systems or organizations. For this reason, it is often stressful for such a cell to exist as a single unit or in isolation of other cells. However, the living environment of the cell could be enriched biochemically or made more physically and / or biochemically conducive. This artificial method of propagation could be in the form of micropropagation or macropropagation.

Micropropagation: This is an asexual means of propagation or multiplication of organisms, especially plants and occasionally animals. It involves the use of *in vitro* technology for propagation and/or multiplication of cells, tissues, organs or even whole organisms in artificial media. The material (termed explants) to be propagated in tissue culture is first excised from source, cleaned and disinfected before immersing or introduction to a liquid, semi-liquid or semi-solid medium which is usually enriched with artificial nutrients and hormones as the case may require. The nutrients with which the medium is enriched include macro- and micro-elements assembled in the appropriate concentration. These nutrients are dissolved in the medium together with various hormones in proportions that would facilitate desired developments in the explants. See Fig. 25.

Fig. 25: A plantain plantation (top left) could be entirely diseased (bottom left) and could be replaced with plants regenerated from a callus from a single healthy plant through tissue culture (right).

Forms of Micropropagation: These are: i) Anther and pollen culture as well as *in vitro* androgenesis used for the development of haploid plants; ii) Shoot culture, which involves *in vitro* cultivation of shoot buds; iii) Root culture, in which root tips are propagated *in vitro*; iv) Protoplast culture, that involves *in vitro* cultivation of protoplasts. Such protoplasts can be used to regenerate whole plants or even used for transformation; v) Somatic culture in which a vegetative part of a plant, be it leaf, cortex, pith tissue, etc. is cultured; vi) Callus culture in which an explant is induced to grow into callus (i.e. a mass of undifferentiated thin-walled

cells); vii) Embryo culture is *in vitro* propagation of an embryo. It is often treated as a form of somatic tissue or organ.

Others are: viii) Embryogenic culture which is a type of culture in which embryo-like tissues are derived from somatic tissues and calli in culture; ix) Embryo rescue, which is used to salvage embryos formed from crosses in which the endosperm could not sustain the embryo. Such embryos are excised and cultivated *in vitro* (e.g. plantain and banana hybrids lack viable endosperm due to a form of cytogenetic failure and require embryo rescue to be successfully propagated); x) There is cryo-preservation whose sole purpose is for storage of living organisms such as plants in ice for as long as they are so-desired. The materials in cryo-preservation could be reactivated back to life any time such is desired. Unlike routine breeding procedures, products of micropropagation require acclimatization and/or hardening in order to gain the ability to live normally.

Macropropagation: This is asexual multiplication of plants *in vivo*. In this technology, a plant is treated in a way that would induce enhanced adventitious meristematic activity. This would induce the lateral buds or adventitious buds to start rapid activity. Usually, the meristematic activity leads to development of several adventitious buds growing into branches or propagules. In bananas, plantains and pineapples (Agogbua and Osuji, 2011), this process leads to production of suckers that develop and grow according to age. See Fig. 26.

This way all the buds develop into suckers so that one plant material could yield dozens of planting materials. Macropropagation offers an ideal method for mass-production of genetically transformed plant materials. One advantage of macropropagation is that where the genetic transformation or

transgenization is not uniform in all the target cells, chimera could form, and as such may offer biodiversity.

Fig. 26: Split crown technique of mass producing Sweet Cayenne pineapple.

Encapsulated or Artificial Seeds: Somatic embryos (i.e. products of somatic embryogenesis) can be excised and covered or encapsulated in a protective solid water soluble gel (i.e. hydrogel). The solid gel is constituted of sodium or calcium alginate and impregnated with basic nutrients and therefore serve the purpose of both endosperm and seed coat. The alginate is a product of brown algae. The somatic embryo remains protected in the encapsulating medium but when planted in the soil, the alginate absorbs water. Absorption of water by the alginate enables it to release the nutrients

contained in it to the somatic embryo which in turn starts to grow in the pattern of seed germination.

APPLICATIONS OF MOLECULAR GENETICS IN BIOTECHNOLOGY

Respected audience, biology has become extraordinarily exciting over the past few decades. Due to few strategic discoveries, we are now in an era of biology termed 'biotechnology'. The advances and achievements of this wonderful era could not have been envisaged about twenty to thirty years ago. A wide range of organisms and their constituents are being exploited to fulfil more needs for the farming, food, pharmaceutical and chemical industries. The biotechnology practiced today has already laid a strong foundation for very significant future milestones.

Recent progress in molecular genetics enables us to isolate genes and to determine how genetic information gives rise to useful characteristics of the whole organism. In addition, it is now possible to understand in detail the complex biochemical circuitry that lies between the genetic blueprint and the development of a particular feature. This understanding enables us modify the genetic information in ways that will create variations, or introduce new and favourable characteristics in organisms.

Technical and intellectual innovations responsible for the progress in molecular genetics have, in recent times, attracted scientists in the academia and industry who continually contribute new and novel ideas towards continued development of biotechnology. More competitive culture for research and development has given rise to new discoveries, which in their totality have revealed that many principles, processes and mechanisms are common to all organisms. For

instance, the centromeric and telomeric DNA sequences are ubiquitous. For this reason, the centromere and telomere of several plant and animal chromosomes are interchangeable.

The telomere-like sequences of wheat, barley, and some other plants are similar to that found in bananas and plantains (Osuji *et al*., 1998a). In the same way, ribosomal RNA genes otherwise known as ribosomal rDNA is similar between several plant families including Musaceae, Cucurbitaceae, Poacea, etc. (Osuji, *et al*., 1998a; Osuji, *et al*., 2006) and several animal families. Apart from specific sequences, members of related species and genera share a good percentage of similarity in the genomic DNA (Osuji *et al.,* 1997d). It has been shown that plants and animals share some retrotransposonts (Balint-Kurti *et al*., 2000; Baurens *et al*., 1997).

Research in biotechnology and its exploitation is increasingly competitive. More scientists are becoming interested. Increased participation by the industry globally is leading to more investments. The end-products of biotechnological research are becoming more evidently unlimited and cutting across major interests and interest groups. As investment increases worldwide, and new applications emerge, the global public are increasingly becoming aware of the problems and benefits coming from biotechnology.

Within the scope of biotechnology, molecular genetics techniques have been developed to: i) produce rare and medicinally active molecules, ii) change hereditary traits of plants and animals, iii) diagnose diseases, iv) produce useful chemicals with pharmaceutical implications, v) cleanup and restore environmental health conditions and vi) provide means of discriminating between taxonomic units of life.

Biotechnology has therefore revolutionized i) agriculture, ii) medicine, iii) industry, iv) biodiversity conservation, v) military art of warfare and vi) environmental protection. It has also influenced the engineering industry enormously especially since new expected products require design and composition of new resource equipment, and as well, generate fund for industrial and economic growth.

I am interested in discussing how the entire issue affects man's hunger for food, his desire to subdue poverty and enjoy good health.

War against Hunger

Vice-Chancellor Sir, God commanded man to subdue the earth. Hence man subdued and domesticated some crops and animals. Crops are mainly meant for consumption to subdue hunger and maintain good health. The steady increase and occasional explosion of human population in conjunction with anthropogenic factors such as urbanization, industrialization, environmental pollution and contamination (Nwachukwu and Osuji, 2007), continue to pose threats to the healthy existence of man. Natural processes such as erosion, desertification, floods, rain and wind storms, drought, famine, etc. depreciate available farm resources. See Figs. 27 -30.

Application of biotechnology has helped to solve some of these problems of food production caused by natural disasters and anthropogenic factors as well as increased pressure on available resources. However, application of molecular genetics through genetic manipulations, mutation breeding and biotransformation in combination with biotechnology has helped to address issues bordering on food crises.

These have been achieved through tissue culture; cryo-preservation; maximum production of specific proteins,

alteration of enzymes e.g. to render organisms resistant to pests, parasites and herbicide effects; Silencing of an endogeneous gene; addition of new traits e.g. increased productivity and drought tolerance; control of insect pests and parasites using male sterility genes; creation of new ornamental plants such as flowers with altered petal shapes, colours and textures; etc. Specific examples are treated below as follows:

Production of high yielding crops

Crop selections belonging to cassava, cowpea, potato, maize, wheat, rice, sugar cane, among others have been genetically modified to give higher yield. Such transgenic or genetically modified (GM) crops have shown great potential for improvement of productivity and therefore higher food production. Genetic modification of crops lead to quicker result when compared with products of routine plant breeding.

Apart from incorporation of genes that directly express high yield, genes can also be introduced which confer disease resistance and drought tolerance thereby leading to high survival rate and consequently high productivity. Biofortified cassava has been produced that has better nutritional value and higher protein contents. Such biofortified transgenic cassava was imported into Nigeria and taken to the National Root Crops Research Institute, Umudike for confined field trial.

Production of Disease Resistant Plants

Molecular genetics and biotechnology are also exploited to create crops and farm animals that have high productivity and are disease resistant. This feat is important because disease condition can rubbish high crop and animal production. For example, Panama disease at some time and later Black sigatoka disease brought the productivity of high yielding

plantain and banana estates to zero. Incorporation of disease resistant genes into plants and animals increase their survival potential and enhance their productivity.

For instance cloning of a gene for disease resistance into an orange that has sweet taste, large fruit size, but high susceptibility to disease can dramatically enhance the productivity of such crops. This would lead to a crop with high yield and marginal profit. Genes with products that are toxic to pests and parasites, but not toxic to human consumers, can be incorporated into edible crop plants to improve their resistance to such pests and parasites. There is a Maruca-resistant Cowpea confined field trial at Institute of Agricultural Research 'IAR' Zaria.

Fig. 27: Disease resistant plants; A) Kenyans examining insect-resistant transgenic Bt corn; B) groundnut leaves being

extensively damaged by European corn borer (bottom left image, B1) and Bt-toxins present in groundnut leaves (bottom right image, B2) protect it from extensive damage caused by European corn borer

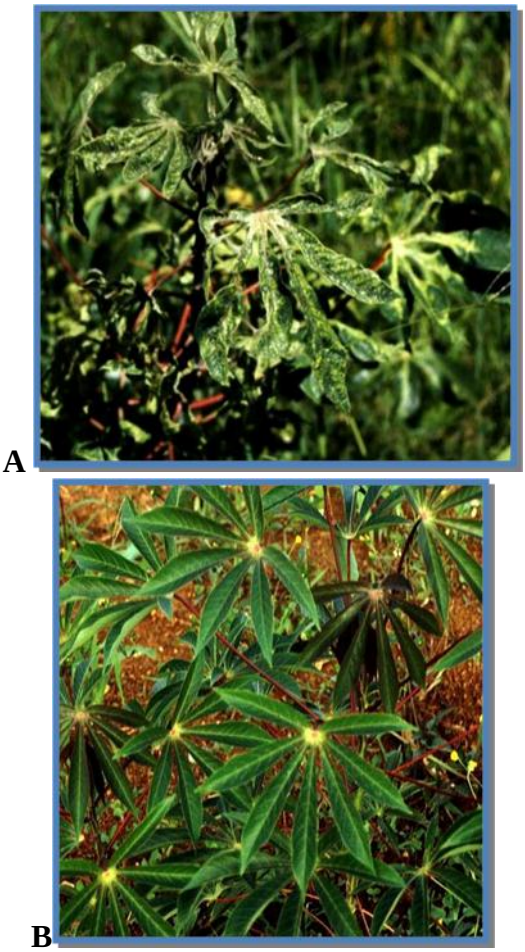


Fig. 28: A) Cassava infested with Cassava Mosaic Virus; B) Normal cassava plant

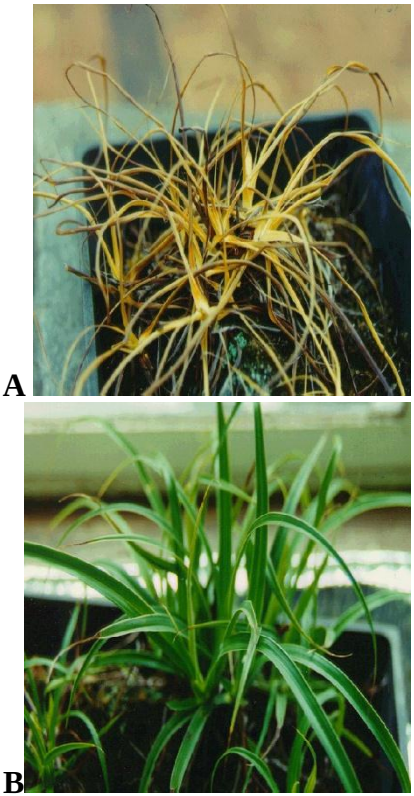


Fig. 29: A) Dehydrated plant lacks the gene for drought tolerance and; B) Drought-tolerant plant (Permission from Prakash)

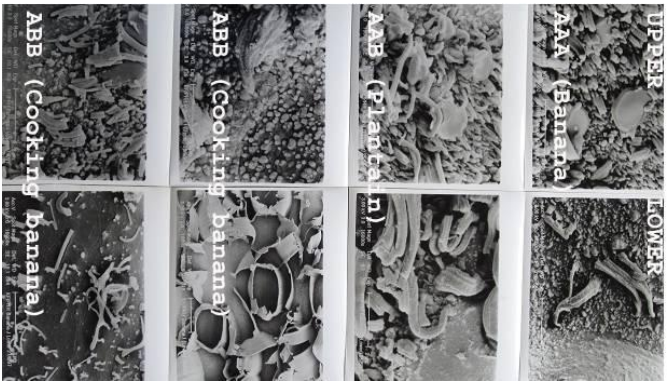


Fig. 30: Gene for resistance to fungal pathogens and gene for drought tolerance could be incorporated into a high-yielding plant to make it disease resistant and drought tolerant as well as high yielding. (With permission from Osuji and Heslop-Harrison, unpublished)

Production of Viral Disease-free Plants

The plantain and banana research has witnessed terrible drawback because of incidences of certain diseases. For instance, the Black Sigatoka disease caused by *Mycosphaerela phijiensis* constituted a huge hindrance to plantain and banana yield and took so much fund to address the problem. One method of solving this problem was by crossing susceptible plants with Black sigatoka-resistant wild diploids. While such crosses yielded relatively black sigatoka-resistant hybrids, the hybrids were discovered to be susceptible to Banana Streak virus (BSV), a badnavirus in the subgroup pararetrovirus.

It was molecular cytogenetics that provided an explanation to the crises (Harper *et al*., 1999). The works of Osuji and coworkers showed that the wild male parental stock that contributed the gene that conveyed black sigatoka resistance had BSV episomatic DNA functionally integrated into its genome. Crossing this material with the female-

contributing banana and plantain cultivars yielded hybrids with genomic BSV infections (Figs 31 and 32). The outcome was wholesale crises in the international banana and plantain research system under the auspices of the International Network for the Improvement of Banana and Plantain 'INIBAP' and Consultative Group on International Agricultural Research 'CGIAR'. Every such hybrid tested was BSV positive, meanwhile most of the hybrids had been injected into national agricultural systems including the Nigerian Agricultural Development Project 'ADP' and the Nigeria Agip Oil Company's (NAOC) Green River Project.

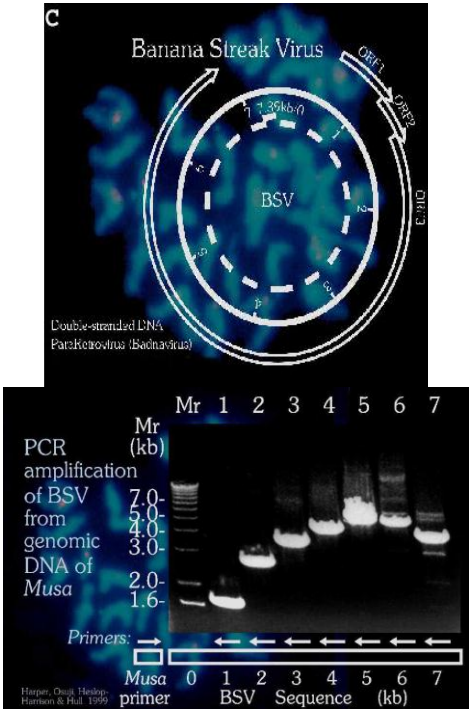


Fig. 31: Result of amplification of BSV DNA from genomic DNA of *Musa*.

Discovery of the imminent BSV epidemic led to mass destruction of plantain and banana estates worldwide. Molecular genetics holds three approaches to the solution of this BSV enigma as follows: i) site-specific mutation by methylation can silence the virus; ii) site specific knockout mutation as has been tried on mice can eliminate the virus or iii) chromosome dissection using laser beams to remove the viral sequence. One other approach could be to search for clean wild male parental stock and start afresh to crossbreed.

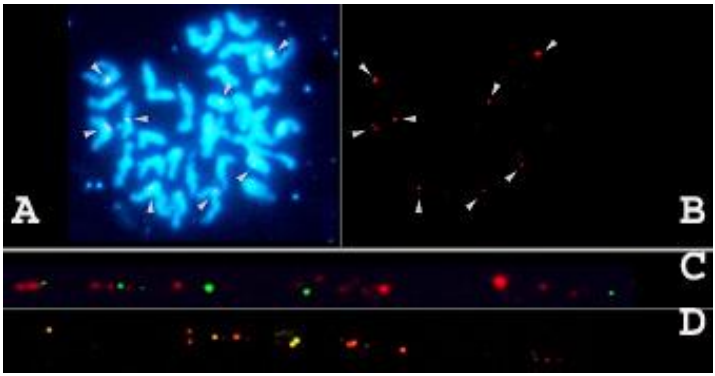


Fig. 32: Micrographs showing A) signals of BSV DNA over-laid on metaphase chromosome spread of triploid banana hybrid, B) the BSV signals shown using a single band width filter, C & D) two different insertion patterns of the BSV DNA in different chromosomes of the banana accession. (Courtesy of Harper, Osuji, Heslop-Harrison and Roger Hull, John Innes Centre, Norwich, England)

Meat and milk production

Novel genes have been incorporated into the genome of several farm animals to impart in them the ability to express various desirable characteristics required to meet goals compliant with the United Nation's MDG. Animals used for

meat and milk production have been highly used for such programmes. See Figs. 33 and 34.

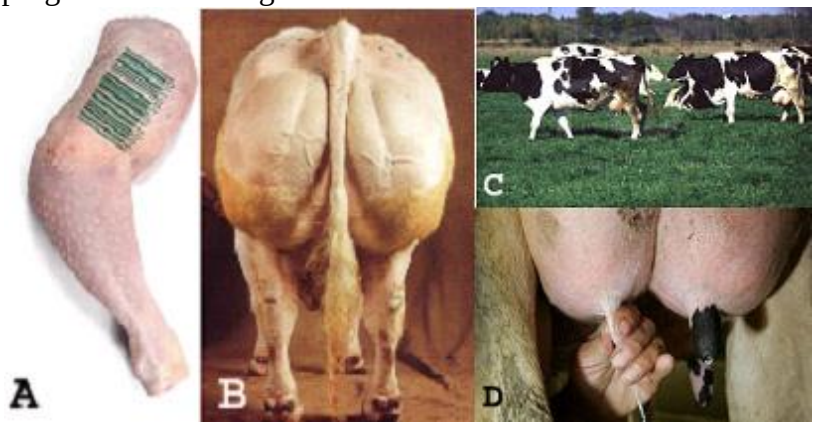


Fig. 33: A) Fast growth, early maturity chicken (1 month maturity) and high meat yield chicken; B) Belgian Blue Bull used for crossing for high vigour and beef yield [Eenennaam, 2008]; C and D) Dairy cows: Vermont green mountain state cows, Jersy.

Fig. 34: Stock Photography - England, North Yorkshire, Harrogate, Dairy cattle awaiting judging at the Great Yorkshire Show

War Against Disease

In recent times, biotechnological developments have closed or narrowed the gap between pharmaceutical and biotechnological industries. Chemical drugs are being rapidly replaced by generic drugs and other products of biotechnology. For example, biotechnologically derived proteins and polypeptides form a new class of potential drugs (Table 3). These products (or molecules) replace lacking or misfunctional equivalents in the body. Insulin, for instance, was formerly extracted from slaughter animals and used for treatment of diabetics. But since 1982, human insulin (Humulin®) has been produced by micro-organisms in fermenters and used for treatment purpose.

Table. 3: Products synthesized by Genetic Engineering

Peptide or Protein	**Potential Use**
α_1-antitrypsin	Treatment of emphasema
α-,β-, and γ-interferons	As antiviral, antitumor, anti-inflammatory agents
Blood clotting factor VIII	Treatment of hemophilia
Calcitonin	Treatment of osteomalacia
Epidermal growth factor	Treatment of wounds
Erythropoetin	Treatment of anemia
Growth hormone	Growth promotion
Insulin	Treatment of diabetes
Interleukins-1, 2 and 3	Treatment of immune Disorders and tumours
Macrophage colony Stimulating factor	Cancer treatment
Relaxin	Aid to childbirth
Serum albumin	Plasma supplement
Somatostatin	Treatment of acromegaly

Streptokinase	Anticoagulant
Tissue plasminogen activator	Anticoagulant
Tumour necrosis factor	Cancer treatment

Other Molecular Genetics-facilitated Biotechnologically produced drugs

Similarly, hepatitis B vaccines such as RecomBivax HB® (from Merk), Guni® (from Shantha Biotechniques Limited, Hyderabad), Shanvac® (Biological E. Laboratory) etc. are biotechnologically produced. Production and demand for protein drugs have risen considerably. Generic drugs which include nucleic acids are also being used. The hormone Bovine Somatotropin 'BSTC' is administered to cows to increase milk yield. This and other forms of Molecular pharming are on the increase.

Preparation of most of these drugs were aided by molecular genetics in the sense that genomic domains containing the functional genes were mapped, DNA strands containing the genic sequences were obtained and cloned (through transformation) into cells of other organisms. Furthermore, cellular or extracellular products of such transformed cells were isolated, collected, purified and processed to yield the functional products that are applied as drugs.

Recently in China, genetically modified cows have been produced that produce human milk (Fig. 35). This breakthrough is targeted at producing commercially affordable human milk for babies whose mothers are either unable to breast-feed or those whose mothers are diseased. This raises hope for survival of such babies. Similarly, cows with human immunoglobulins have been produced, which may provide an important source of polyclonal antibodies for the treatment of a

variety of medical conditions including organ transplant rejection, cancer, and autoimmune diseases, such as rheumatoid arthritis.

Fig. 35: A) Human milk cow: Chinese scientists genetically engineered 300 dairy cows to produce milk that contains nutrients found in human breast milk; B) Genetically engineered cows producing human immunoglobulins (Hematech, Sioux Falls, S.D.) (Photo by Alison Van Eenennaam, University of California, Davis)

With the aid of molecular genetics, recombinant DNA technology has led to the production of several other forms of

transgenic animals, which are meant for various human health-related conditions. See Fig. 36 below.

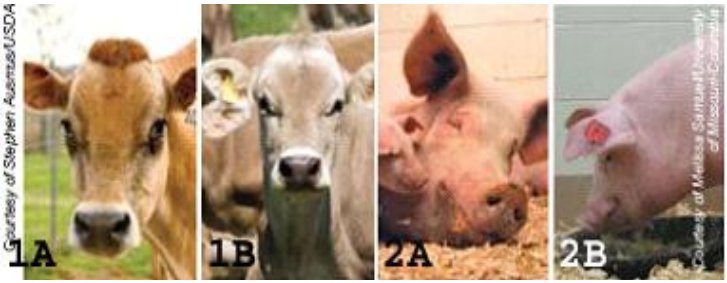


Fig. 36: The US Department of Agriculture's Agricultural Research Service genetically engineered the cow (1A) to produce an antimicrobial protein to resist infection with *Staphylococcus aureus* and Researchers at the University of Missouri-Columbia genetically engineered the pig (2B) to serve as a model of cystic fibrosis in humans. 1B and 2A are controls.

Nutraceuticals

Nutraceuticals are products, which serve both as food and as drugs. Such products are established by incorporating a gene for the production of a medicinally active compound into a food crop. The crop product when harvested would contain both food nutrients and the medicinal compound. Hence when consumed, such a food item would equally play the role of a drug. An example is incorporation of insulin gene in a fruit crop like garden egg. If the transgenic garden egg produces fruits that contain insulin, the fruits can serve both as snack and as a medicine for diabetes. One essence of establishing nutraceuticals is to add more value to food crops

Facilitation of Gene Therapy

Gene therapy is a novel pharmacological approach in which the drug is supplied in the form of a nucleic acid – DNA, RNA, or some modification or combination thereof. Molecular genetics facilitates gene therapy by aiding recombinant DNA technology i.e. genetic engineering. The gene of interest has to be first identified and mapped before it can be cut out for introduction into a recipient cell or organism. Appropriate molecular genetics steps ensure that the right gene is isolated for modification and/or amplification for use in transformation.

Usually the transformed or genetically engineered cells are marked by attachment of a marker gene (which confers a phenotypic characteristic) to the gene of major interest before recombination. The purpose of the marker gene is to enable identification of cells that are successfully transformed.

Using Molecular Genetics Techniques to investigate Human Diseases

Genome analysis is important for medical diagnosis and therapy (Figs. 37 and 38). For instance, it is used for screening specific gene mutations or genetic factors determining the susceptibility to certain diseases; for tracking chromosomal aberrations and other chromosomal anomalies that lead to disease conditions (Table 4); for identifying bacterial and viral infections; for the development of vaccines, and for gene therapy.

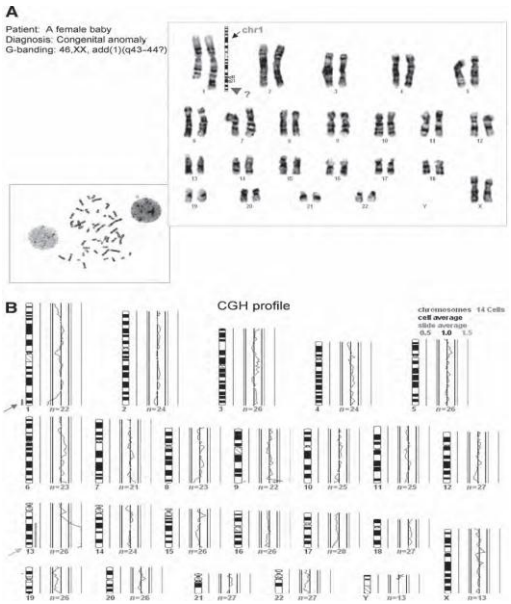


Fig. 37: The application of molecular cytogenetics tools in detecting the chromosome aberrations in a case with congenital anomaly. (A) Traditional G-banding analysis finds an abnormal chromosome with additional materials in the terminal end of one chromosome 1q. However, the origin of this material is unable to be determined by G-banding. (B) CGH [Comparative Genomic Hybridization] is then performed and shows genomic changes with 1q terminal loss and 13q gain. (C) FISH [Fluorescent *in situ* Hybridization] with chromosome painting probes 5 and 13 (chromosome 5 painting probe is used as an internal control) is performed and shows the additional material in chromosome 1q terminal is from chromosome 13q. (D) FISH with 13q and 1q telomere probes is also done and detects the deletion of terminal region in one of chromosome 1. All of these FISH studies confirm the finding by CGH analysis and give us a clearer picture about this chromosome aberration

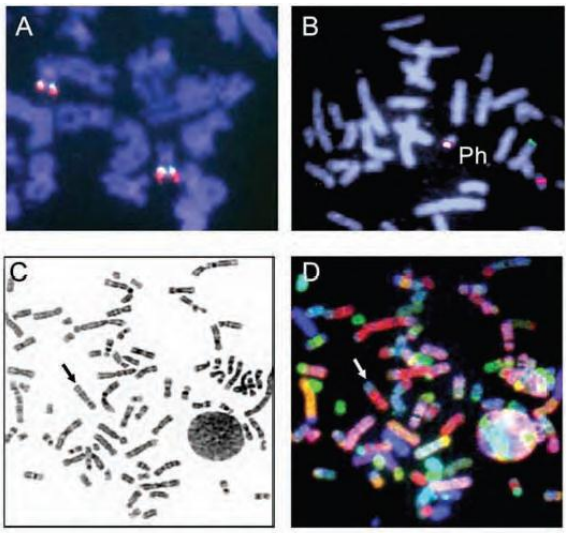


Fig. 38: Examples of Metaphase and Interphase FISH analysis from Cytogenetic preparations. A). Mapping of BAC clones to human chromosome 19 showing the mapping location of two labeled BAC clones (red and green) that co-localized to the expected 19 region of the human genome. B). Example of the Ph chromosome in a CML specimen. Shown are the results of metaphase FISH analysis using probes for the BCR gene located on chromosome 22 (red) and for the ABL gene on 9 (green); C–D). Example of SKY analysis on a human ovarian primary tumor. In C, the DAPI counterstaining has been inverted to mimic the conventional banding patterns of chromosomes enabling identification. D). shows the hybridization of the 24-color probe cocktail to the metaphase spread and reveals that in addition to the excess of chromosomes over the normal 46, numerous chromosomal rearrangements are present and recognized by the change of color along the length of a continuous chromosome. In a normal karyotype, each chromosome possesses a specific

fluorescence uniform signature, along the length of the chromosome (Fig. 38).

Table. 4: Examples of Common and/or important conditions that can be examined using Molecular Cytogenetic Methods:

Medical conditions	Medical conditions continue.
Common chromosome anomalies • Down's syndrome. • Turner's syndrome. • Klinefelter's syndrome. • Translocations.	***Single gene disorders*** ***Autosomal dominant disorders*** • Adult polycystic kidney disease. • Neurofibromatosis. • Huntington's disease. • Hypercholesterolemia.
Recessive disorders • Cystic fibrosis. •Haemoglobinopathies (sickle-cell disease, thalassaemias). • Haemochromatosis.	***X-linked disorders*** • Duchenne and Becker muscular dystrophies. • Haemophilia A. • Fragile X.
Multifactorial diseases • Familial forms of common diseases (e.g. breast cancer, bowel cancer). • Disorders with a genetic component (e.g. cerebrovascular disease, cardiovascular disease, Alzheimer's, asthma).	***Familial cancers*** • Breast. • Colon.
Conditions exhibiting variable inheritance patterns • Inherited forms of deafness. • Muscular dystrophies.	

War Against Poverty

Molecular genetics has facilitated biotechnology in such a way that what is now known as modern day biotechnology is driven by recombinant DNA technology i.e. Genetic engineering which in turn is driven by molecular genetics. A successful transformation or successful creation of a new or modified organism first of all attracts fund through patent. Commercialization of the new product yields more fund not only to the scientists and laboratories involved but also to downstream actors such as subsistent farmers and traders. Several countries have recently invested huge amounts of money in molecular genetics-driven modern biotechnology.

For instance, the Nasdaq Biotechnology Index rose 457% from the end of August 1998 to the end of February 2000. Going back even further to the early 1990s, biotech stocks had soared by 1,347%. For biotech investors every $10,000 invested turned into nearly $140,000. The good news for investors is that after slumping during the recession, biotech stocks have made a comeback. In the first quarter of 2012 alone, the Nasdaq Biotech Index gained 18.2% and conditions are setting for better gains in the future (Miller, 2012)

The United States of America in 2001 alone invested heavily on crops - soybeans, corn, canola and cotton, and in 2008 alone added 9.4 billion dollars to farmers. The global biotechnology industry consists of about: 1,500 companies with Annual industry revenues of USD 40 billion and market capitalization exceeding USD 300 billion. In India industry size in 2002 was estimated at US$ 2.5 billion. This implies that the industry size as at 2012 would have increased significantly. During the period 1996-2008, Biotech crops reduced pesticide spraying by 352 million kg (-8.4%) and as a result decreased the negative environmental impact associated with herbicide

and insecticide use on the area planted with biotech crops by 16.3%. This additional positive side of biotechnology underscores the need for its continued patronage. Naturally, that type of activity has caught the eye of the markets, as biotech has suddenly become the hot sector of late as money rotates out of commodities (Christ, 2008). See Fig. 39.

Countries like Brazil and India are already making progress out of the unfortunate club of third world countries. Hence they have turned into emerging economies due to their concerted effort to increase per capita income through biotechnology. South Africa has also joined, as a frontline African nation, and has started making exports of biotechnology products. Presently, Kenya earns huge foreign exchange from exportation of biotechnology-aided horticultural products.

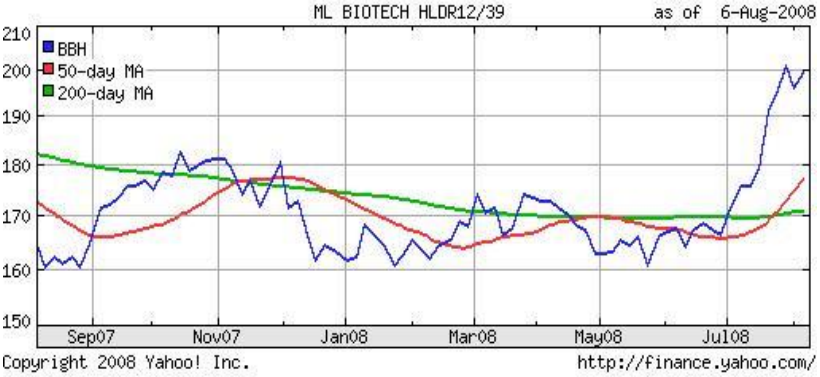


Fig. 39: Bullish on Biotech Investments as biotech has suddenly become the hot sector of late as money rotates out of commodities (Christ, 2008).

Where does Nigeria Stand in the Picture?

Modern biotechnology regulation in Nigeria started in the early 1990s. The Nigerian 'Biosafety Guidelines' was developed in 1994 and it covered only agricultural biotechnology. However, it took place before the Convention on Biodiversity 'CBD', and the Cartagena Protocol on Biosafety (2000, 2002, 2003). Development of more encompassing Biosafety Guidelines in line with the Convention on biodiversity took place in 2001 to encourage research, development and regulation of modern biotechnology. However, it was not binding on any interested party. Inauguration of the National Coordinating Committee (NCC) for development of a bill on biosafety took place in 2002.

The National Biosafety Framework (NBF) was established and The National Coordinating Committee (NCC) submitted its report on NBF on 2nd December 2002, with the following components: i) National biosafety policy, ii) Biosafety draft bill, iii) Public awareness and participation in biosafety and iv) Request and authorization. After due but prolonged process, the National Biosafety Bill has been put in place, which i) covers all genetically modified organisms/living modified organisms, products, food/feed and processing; ii) confers the power to carry out risk assessment and risk management; iii) defines offences and Penalty for violation of the act; iv) covers socio-economic consideration in risk assessment, liability and redress; and v) Labeling of all GM products, etc.

Despite this initial effort, Nigeria has not made any significant move to join the global biotechnology trend. Following a bill that was sent to the House of Assembly, a study tour of the Phillipines GM crop Farms was organised for the House Committees on Agriculture, Environment and

Science and Technology to have a practical experience on development of GMOs and how they are being regulated as well as the legislation procedure. I guess the tour was in line with the Nigerian spirit.

National Biotechnology Development Agency (NABDA) was established to anchor development of biotechnology in the country. However, it is not evident that Nigeria's geopolitics has not undermined NABDA's mandate. National Centre for Genetic Research and Biotechnology (NACGRAB) was also established at Ibadan. What is largely unclear is if these establishments were to be financially autonomous or to draw fund from government sources. Meanwhile, the so called Biotechnology establishments are not being controlled by Molecular Geneticists, Molecular Biologists, Biochemists or career biotechnologists.

Despite all these institutional frameworks to support Biotechnology development in Nigeria, both the public and private sectors of the country's economy have not made any significant effort to sustain applied biotechnology. For now, imported biofortified transgenic cassava is undergoing 'Confined Field Trial at the National Root Crops Research institute (NRCRI), Umudike. The Federal Government also gave permit for construction of enabling facility and later for Confined Field Trial of Maruca-resistant cowpea. The International Institute of Tropical Agriculture (IITA) conducted biotechnological researches in the area of crop-based agriculture. I would like to state at this juncture that the IITA researches were obviously restricted to the institute's mandate crops and would not serve any strategic national interest for Nigeria. However, Nigeria needs to establish and position her own biotechnological industry if she is interested in economic and socio-political advancement.

Where Does the University of Port Harcourt come into the picture?

Basically, the first step in establishing biotechnology capability is in having staff trained in the profession. Secondly, there is need for fund raising using every possible means to establish the enabling infrastructure. Thirdly, there is need for commitment to the cause and to mobilize institutional support for research by creating locally enabling environment. Staff in all fields, who show adequate commitment to research, should be sponsored to International conferences so that they can follow the trend of research in the global community. It is unfortunate to state here that most of the researches conducted by our academics are old fashioned and redundant. People here are researching in the areas the world has left behind. Consequently, most of our research findings make no sense to the global community. Once there is a will, there would be a way for improvement.

Social Impacts of Molecular Genetics-Driven Biotechnology

Biotechnology as facilitated by molecular genetics has several benefits, which include reduction of hunger, poverty, disease and death. However, it has several risks. Several chemicals and reagents used in molecular genetics and biotechnology are environmentally toxic while some others are capable of causing direct health hazards. It is therefore imperative to conduct careful analysis of risks and benefits to avoid problems in the process of establishing a brotec facility.

Safety Considerations

Safety protocols should be put in place to avoid: i) widespread infections caused by genetically engineered microorganisms

(GEMs), ii) spread of genes from GEMs to other microorganisms in the environment and iii) release of GEMs currently regulated by several federal agencies.

Ethical and Moral Considerations

Ethical and moral standards governing sensitive scientific research should be considered such as prohibition of: i) genetic engineering of humans, ii) unethical use of genetic information obtained from an individual and iii) creation of biological weapons or bioterrorism.

Environmental Considerations

Access of dangerous reagents and chemicals (which have the capability of dissolving and draining down the soil) to ground water. This situation should be checked by putting in place proper waste management structures especially to check: i) ecosystem disruption, ii) spread of cloned genes to weeds or other organisms in the environment and iii) access of dangerous chemicals and reagents to water supply systems.

Conclusion

Vice-Chancellor Sir, what evolution does naturally is the same thing we do through genetic engineering. Hence, the more we look into organisms, the more we see characters. The more we look into characters, the more we see genetics. The more we look into genetics, the more we see cytogenetics. The more we look into cytogenetics, the more we see molecular genetics. The more we look into molecular genetics, the more we see genomes. The more we look into genomes, the more we see genes. The more we manipulate genes, the more new generic products or modified organisms emerge and the more biotechnology develops. The more biotechnology develops the more improved productivity and yield, disease resistance,

drought tolerant and environmentally stable organisms emerge. The more these outputs of biotechnology emerge, the more richer and healthier individuals and corporate groups emerge. The more richer individuals, organizations and nations emerge, the more poverty disappears. The more poverty disappears, the happier we become. Thank you for listening.

References

Agogbua, J.U. and **Osuji, J.O.** 2011. Split Crown Technique for Mass Propagation of 'Smooth Cayenne' Pineapple in South-South Nigeria. *African Journal of Plant Science*. 5 (10): 591-598.

Anyanwu, D.I. and **Osuji, J.O.** 2001. An evaluation of the agroforestry capability of *Vigna unguiculata* (L.) Walp. Under attenuated light conditions. *Journal of Innovations in Life Sciences*. 6(2): 17-24.

Anyanwu, D.I. and **Osuji, J. O.** 2002. Effect of light attenuation on the chlorophyll content of four varieties of *Vigna unguiculata* (L.) Walp. *African Journal of Applied Zoology and Environmental Biology*. 4: 12-16.

Balint-Kurti PJ, Clendennen SK, Dolezelovca M, Valarik M, Dolezel J, Beetham PR, May GD. 2000. Identification and chromosomal localization of the monkey retrotransposon in *Musa* sp. *Molecular and General Genetics* 263: 908–915.

Baurens FC, Noyer JL, Lanaud C, Lagoda P. 1996. Use of competitive PCR to assay copy number of repetitive elements in banana. *Molecular and General Genetics* 253: 57–64.

Baurens FC, Noyer JL, Lanaud C, Lagoda P. 1997. Copia-like elements in banana. *Journal of Genetics and Breeding* 51: 135–142.

Bennett MD, Leitch IJ. 2004. Plant DNA C-values database (release 3.0, Dec. 2004). http://www.rbgkew.org.uk/cval/homepage.html. Accessed July 2007

Biotechnology Industry Organization 'BIO' 2007. The Guide to Biotechnology, 2007. Biotechnology Industry Organization.

Browne, Janet. 1996. *Charles Darwin.* Princeton, NJ: Princeton University Press.

Buhariwalla HK, Jarret RL, Jayashree B, Crouch JH, Ortiz R. 2005. Isolation and characterization of microsatellite markers from *Musa balbisiana*. *Molecular Ecology Notes* 5: 327–330.

Carreel F, Gonzalez de Leon D, Lagoda P, Lanaud C, Jenny C, Horry J, Tezenas du Montcel H. 2002. Ascertaining maternal and paternal lineage within Musa by chloroplast and mitochondrial DNA RFLP analyses. *Genome* 45: 679–692.

Charlesworth, B., Sniegowski, P. and Stephan, W. 1994. The evolutionary dynamics of repetitive DNA in eukaryotes. *Nature* 371, 215–220.

Christ, S. (2008). Biotech Investments- The Brewing Bull Market in Biotech. WealthDaily

Cooper, G.M. 2000. *The Cell: A Molecular Approach*. 2nd ed. ASM Press, Washington D.C., Sinaur Associates, Inc., Sunderland, Massachustts.

Crick, Francis. 1988. *What Mad Pursuit?* New York: Basic Books,

Dover, G.A. 1982. Molecular drive: A cohesive mode of species evolution. *Nature* 299, 111–117.

Dubey, R.C 2004. *A Textbook of Biotechnology*. S. Chand & Company Ltd., Ramnagar, New Delhi-110 055.

Eenennaam, E.V. 2008. *Genetically Engineered Animals: An Overview*. UC Davis. http://animalscience.ucdavis.edu/animalbiotech

Ekanem, A.M. and **Osuji, J. O.** 2006. Mitotic index studies of edible cocoyams (*Xanthosoma* and *Colocasia*). *African Journal of Biotechnology*. 5(10): 846-849.

Evans, G.M. and Furlong, J.C. 2003. *Environmental Biotechnology: Theory and Application*. John Wiley &

Sons, Limited., The Atrium, Southern Gate, Chichester, West Sussex PO19 8SQ, England.

Faure S, Noyer J-L, Carreel F, Horry J-P, Bakry F, Lanaud C. 1994. Maternal inheritance of chloroplast genome and paternal inheritance of mitochondrial genome in bananas (Musa acuminata). *Current Genetics* 25: 265–269.

Finch, J.T. and Klug, A. 1976. Higher order of chromatin structure. *Proc. Natl. Acad. Sci.* USA. 73: 1900.

Harper, G., **Osuji, J. O.,** Heslop-Harrison, J.S. and Hull, R. 1998c. Integration of banana streak badnavirus into the *Musa* genome: molecular and cytogenetic evidence. *Virology*, 255: 207-213.

Hartl, Daniel L., and Elizabeth W. Jones. 1998. *Genetics: Principles and Analysis*, 4th ed. Sudbury, MA: Jones and Bartlett.

Hartwell, L.H., Hood, L., Goldberg, M.L., Reynolds, A.E., Silver, L.M. and Veres, R.C. 2000. *Genetics: From Genes to Genomes*. McGraw-Hill Higher Education, Boston Burr Ridge, Il Dubuque, IA Madison.

Heslop-Harrison JS. 1991. The molecular cytogenetics of plants. *Journal of Cell Science* 100: 15–21.

Heslop-Harrison, J.S. 2000. Comparative genome organization in Plants: From sequence and markers to chromatin and chromosomes. *The Plant Cell*. 12: 617-635.

Heslop-Harrison JS, Brandes A, Taketa S, Schmidt T, Vershinin AV, Alkhimova EG, et al 1997. The chromosomal distributions of Ty1-copia group retrotransposable elements in higher plants and their implications for genome evolution. *Genetica* 100: 197–204.

Heslop-Harrison, J.S. and Schwarzacher, T. 2012. Domestication, Genomics and the future of Banana. *Annal of Botany*, 100: 1073-1084.

Heslop-Harrison, J.S., **Osuji, J.**, Hull, R. and Harper, G. 1999. *Fluorescent In situ Hybridization of Plant Chromosomes: Illuminating the Musa Genome*. INIBAP Annual Report 1998. INIBAP, Montpellier, France,p. 26-29.

Heslop-Harrison, J.S. and Schwarzacher, T. 2007. Domestication, genomics and the future for Banana. *Annals of Botany*, 100: 1073-1084.

Kornberg, R.D. 1974. Chromatin structure: A repeating unit of histones and DNA. *Science*: 184: 868-871.

Larson, E.J. 1997. *Summer for the Gods.* New York: Basic Books, *National Center for Science Education.* http://www.natcenscied.org.

Lavergne, S., Muenke1, N.J. and Molofsky, J. 2010. Genome size reduction can trigger rapid phenotypic evolution in invasive plants. *Annals of Botany*, 105: 109–116.

Lewis, R., Gaffin, D., Hoefnagels, M. and Parker, B. 2002. Life. 4th Edition. McGrawHill Publishers, Bostn Burr Ridge.

Mader, S.S. 1998. *Biology,* 6th ed. Boston: McGraw-Hill

Margulis, L. 1998. *Symbiotic Planet: A New Look at Evolution.* New York: Basic Books,

Meagher, T.R., Gillies, A.C.M. and Costich, D.E. 2005. Genome size, quantitative genetics and the genomic basis for flower size evolution in *Silene latifolia*. *Annals of Botany*, 95: 247–254.

Miller, D. 2012. *Biotech Stock ETFs: How to Ride the Surge in Biotech Mergers and Acquisitions*. Money Morning Investor Reports. Article Index FAQ

Müler, S. ed. 2008. *Nucleic Acids from A to Z: A Concise Encyclopedia.* WILEY-VCH Verlag GmbH & Co. KGaA, Weinheim.

Nill, K.R. 2002. *Glossary of Biotechnology Terms*. 3rd Ed. CRC Press, Boca Raton, London, New York, Washinton DC.

Nwachukwu, E.O. and **Osuji, J. O.** 2007. Bioremedial degradation of some herbicides by indigenous white rot fungus, Lentinus subdunus. *Journal of Plant Science*. 2(6): 619-624..

Nwachukwu, E.O. and **Osuji, J. O.** 2008. Evaluation of plant extracts for antifungal activity against S*clerotium rolfsii* causing cocoyam cormel rot in storage. *Research Journal of Agriculture and Biological Sciences*, 4(6): 784-787.

Nyananyo, B.L. and **Osuji, J. O.** 2007. Biosystematic characterization of *Sphenostylis stenocarpa* (Hochst *ex* A. Rich.) Harms. *Nigerian Journal of Botany*, 20(2): 411-419.

OECD. 1989. Document No. 237.94. *Organization for Economic Cooperation and Development*, Paris.

Onyeachusim, H.D., **Osuji, J. O.** and Okoli, B.E. 2006. Distribution and habitats of some endangered rare Nigerian tree species. *Journal of the Nigerian Environmental Society*. 5 (3): 242-248.

Okoli, B.E. and **Osuji, J. O.** 2008. The status of research on the cytogenetics of Nigerian fruit crops. *Nigerian Journal of Botany*, 21(2): 358-372.

Osuji, J.O., Okoli, B.E. and Ortiz, R. 1996a. An improved procedure for mitotic studies of the *Eumusa* section of the genus *Musa* L. (Musaceae). *Infomusa* 5(1): 12-14.

Osuji, J.O., Okoli, B.E. and Ortiz, R. 1996b. Cytological evaluation of plantains, bananas and their hybrids: technique for mitotic study. *Nigerian Journal of Genetics*. 10: 1-7.

Osuji, J.O., Okoli, B.E. and Ortiz, R. 1996c. Conservation and characterization of *Musa* genetic resources. *Nigerian Journal of Genetics*. 10: 8-13.

Osuji, J.O., Okoli, B.E., Vuylsteke, D. and Ortiz, R. 1997a. Multivariate pattern of quantitative trait variation in triploid banana and plantain germplasm. *Scientia Horticulturae*, 71: 197-202.

Osuji, J.O., Ortiz, R. and Vuylsteke, D. 1997b. Ploidy variation in hybrids from interploid 3*x* X 2*x* cross in *Musa*. *Tropicultura*, 15(1): 37-39.

Osuji, J.O., Okoli, B.E. and Ortiz, R. 1997c. Histochemical localization of calcium oxalate crystals in fruits of plantain and banana cultivars. *Fruits*, 52(1): 5-10.

Osuji, J.O., Harrison, G., Crouch, J. and Heslop-Harrison, J.S. 1997d. Identification of the genomic constitution of *Musa* L. lines (bananas, plantains and hybrids) using molecular cytogenetics. *Annals of Botany*, 80(6): 787-793.

Osuji, J.O. 1998. Chromosomal investigations of infra-specific taxa in *Musa paradisiacal* L. (Plantain; *Musa* spp. AAB group). *Journal of Innovations in Life Sciences*, 3: 87-92.

Osuji, J.O., Crouch, J., Harrison, G. and Heslop-Harrison, J.S. 1998a. Molecular cytogenetics of *Musa* L. species, cultivars and artificial hybrids: location of 18S-5.8S-25S and 5S rDNA and telomere-like sequences. *Annals of Botany*, 82: 243-248.

Osuji, J.O., Onyeike, E.N. and Nwachukwu, A.S. 1998b. Histochemical localization of tannins in ripe and unripe fruits of *Musa* L. (Musaceae). *Journal of Innovations in Life Sciences*, 3: 42-47.

Osuji, J.O., Okoli, B.E. and Ortiz, R. 2000. Taxonomic value of calcium oxalate crystals in *Musa* germplasm. *Acta Horticulturae*, 540: 137-146.

Osuji, J.O. and Okoli, B.E. 2001. Enhancing research on plantain, *Musa paradisiaca* L. through molecular cytogenetics. *Nigerian Journal of Genetics.* 16: 1-9.

Osuji, J.O. and Edeoga H.O. 2005. Karyological studies on *Vigna subterranea* (L.) Verdc. from Nigeria. *Cytologia*. 70(2): 167-169.

Osuji, J.O. and Ndukwu, B.C. 2005. Probable functions and remobilization of calcium oxalate in *Musa* L. *African Journal of Biotechnology*, Online. 4(10): 1139-1141.

Osuji, J.O. and Eke, R.O. 2005. Energy pollution and the corrosion of the biosphere and biodiversity. *Nigerian Journal of Corrosion*. 3: 73-77.

Osuji, J.O. 2006. Microstructural characters of the inflorescence bracts distinquish between *Musa sapientum* L. and *M. paradisiaca* L. *International Journal of Botany*, 2(1): 11-16.

Osuji, J.O., Okoli, B.E and Edeoga, H.O. 2006a. Karyotypes of the A and B genomes of *Musa* L. *Cytologia*. 71(1): 21-23.

Osuji, J.O., Okoli, B.E. and Heslop-Harrison, J.S. 2006b. Cytological and molecular cytogenetic studies on *Cucumeropsis mannii* Naudin. *International Journal of Botany*. 2(2): 187-192.

Osuji, J.O. and Nwachukwu, E.O. 2006. Investigations of the occurrence of some heavy metals in the corms of some Nigerian aroid cultivars in the Niger delta region of Southern Nigeria. *Journal of the Nigerian Environmental Society*. 5 (1& 2): 53-57.

Osuji, J.O. and Nsaka, I.J. 2009. Histochemical localization and probable functions of calcium oxalate crystals in Nigerian cocoyams. *Nigerian Journal of Plant Protection*, 26: 91-98.

Osuji, J.O. and Owei, S.D. Jnr. 2010. Mitotic index studies on *Treculia africana* Decne in Nigeria. *Australian Journal of Agricultural Engineering*, 1(1): 25-28

Osuji, J.O. and Agogbua, J.U. 2010. Arrangement of Calcium oxalate structures in the root apex confers a meristem-protection function in yam (*Dioscorea rotundata* Poir). *Scientia Africana;* 9 (1): 166-175.

Padh, H. 1996. *Curr. Sci.* 71: 670-676.

Purves et al., Life: *The Science of Biology*, 4th Edition, by Sinauer Associates

Robinson, R. (ed). 2002. *Biology*. Macmillan Reference, USA, Thomson Gale. Vol. 1. pp. 272.

Sherborn, Victoria. 1995. *James Watson and Francis Crick: Decoding the Secrets of DNA.* Woodbridge, CT: Blackbirch Press.

Strathern, Paul. 1997. *The Big Idea: Crick, Watson, and DNA.* New York: Anchor, Watson, James. *The Double Helix.* New York: Norton Press.

Strahler, Arthur N. 1987. *Science and Earth History: The Evolution/Creation Controversy.* Buffalo, NY: Prometheus Books.

Teuber, V., and Zorn, C. 1982. Rabl's model of the interphase chromosome arrangement tested in Chinese hamster cells by premature chromosome condensation and laser-UV-microbeam experiments. *Human Genetics* 60, 46–56.

CITATION ON
PROFESSOR JULIAN ONYEWUONYEOMA OSUJI, THE 88TH INAUGURAL LECTURER OF THE UNIVERSITY OF PORT HARCOURT

Introduction

It is with pleasure that I stand here today to present the citation on the 88th Inaugural Lecturer of the University of Port Harcourt, in the person of Professor Julian Onyewuonyeoma Osuji, an internationally recognized Molecular Geneticist and Cytotaxonomist.

Born on the 5th of September, 1964 to the family of Ezinna Nze Sir Vincent Benson Oparanwata Osuji and Ezinne Lolo Lady Celestina Osuji of Ogbor Nguru in Aboh Mbaise Local Government Area of Imo State, Professor Julian Osuji is the second child and second son in a family of eight siblings.

Education

As a child, he attended Community School (formerly St. Joseph Primary School), Ogbor Nguru, and had his Secondary education at Community Secondary School (formerly St.

Paul's Secondary School), Ikenanzizi Obowo. Professor Julian Osuji is a pure bred University of Port Harcourt Alumnus, who obtained his B.Sc (Hons.) Botany in 1988, M.Sc. (Biosystematics/Plant Taxonomy) in 1991 and Ph.D. (Biosystematics/Plant Taxonomy) in 1995. His Ph.D. was, however, obtained from the University of Port Harcourt under the studentship of the International Institute of Tropical Agriculture 'IITA'.

Professional Career

On completion of his mandatory National Youths Service in 1989, the young Julian secured admission to study for his M.Sc. degree after which he secured employment as an academic staff of the Alvan Ikoku College of Education, Owerri. After obtaining the Ph.D., he left Alvan for a Postdoctoral Biotechnology Transfer fellowship at the John Inner Centre, Norwich, England. Though he left the Alvan Ikoku College of Education in 1996 as a Lecturer I, he later joined the University of Port Harcourt as a Lecturer II despite the fact that his interview by the University of Port Harcourt for the Lecturer II position was four years earlier (in 1994). The effort he made for proper rank placement in the University yielded no result. He was promoted to the rank of Lecturer I in 2001, Senior Lecturer in 2004. He withdrew his appraisal to the rank of Associate Professor in 2009 because he could not afford any more delay in his professional growth. He was elevated to the Professorial Chair in Molecular Genetics and Cytotaxonomy in the Department of Plant Science and Biotechnology in the University of Port Harcourt in October 2010.

Membership of Professional Bodies

Professor Osuji is a member of the International Society for Horticultural Sciences (in which he is in the Guava Research Group), a member of the Biotechnology Network of Nigeria and a Life Member of International Society of Plant Morphologists. He has served in the National Executive Council of the Genetics Society of Nigeria (2001 – 2003), Nigerian Society for Plant Protection (2005-2011) and currently the Botanical Society of Nigeria (2009 to date).

Administrative Experience

Professor Osuji has had a rewarding public service and academic career with a wealth of experience in Institutional administration. He has served in many statutory and Ad-Hoc Committees of this University, some of which include: Faculty of Science Students' Staff Adviser since 2008; Time Table Coordinator for the Department of Plant Science and Biotechnology from 2005 to 2010; Member, University Committee on Affiliations (2008 – 2010); Member, University of Port Harcourt Senate Committee on Scholarship and Dean's List; and currently Member, Faculty of Science Quality Assurance and Quality Control Committee and Chairman, Faculty of Science Examinations Committee.

Academic Leadership and Publications

Professor Osuji's Academic Leadership has spanned many years of teaching courses in Genetics, Biotechnology and Biosystematics to both undergraduate and postgraduate students. He has written a single author book on research Communication and Presentation. He has also both singly and jointly edited books in his field for both undergraduate and postgraduate programmes. He has authored and co-authored 15 Chapters in 6 Books and has 2 Annual Reports. Professor

Osuji has more than 30 scientific refereed articles 60 % of which was in reputable indexed international journals of which he is lead author in 22. He has delivered 3 Guest Lectures, actively participated in 7 Staff Development Workshops and attended more than 25 Conferences within and outside the country.

He has supervised more than 30 B.Sc. research projects, three M.Sc. theses and currently 1 M.Sc. and 1 Ph.D. dissertations. He has served in the Editorial Board of five Local peer-reviewed journals including Scientia Africana, Nigerian Journal of Genetics, etc. and five international journals namely: International Journal of Botany, Asian Journal of Plant Science, Biotechnology Journal, International Journal of Plant Breeding and Genetics, Research Journal of Botany. He was recently appointed an External Examiner to the Department of Plant Biology and Biotechnology, University of Benin.

Research Interest

His research work adopts a holistic approach to unravel the hidden attributes of the plant life. The work is centered on large scale and long range genome organization of plants. The connection between the genome and the histological and histochemical features and their translation to the external morphology is essentially part of the general research scope. Though, much emphasis is on the karyological domain of the cell and organisms, interaction of environment(s) with gene expression is also an important aspect of his research interest. Major researches he has already conducted cover and are not limited to plant families such as: Cucurbitaceae, Musaceae, Leguminosae (Fabaceae), Araceae and Sterculiaceae on one hand and the vegetations of the Niger delta and rainforest belts of Nigeria on the other.

Professor Osuji was a UNESCO, Gatsby Foundation / IITA Postdoctoral Research Fellow to the John Innes Centre, Norwich, which is a world centre of excellence in Molecular Genetics and Biotechnology in 1996 to 1997 and 1997 to 1998 respectively. He had NUC Research Grants in 2001 and 2002 through the Office of the DVC (Academic) and the World Bank STEP-B Grant for Centre of Excellence in Environmental Protection and Conservation together with the Universities of Maiduguri, University of Nigeria Nsukka and the National Biotechnology Development Agency, Abuja.

National and Community Service

Professor Osuji has served as a Consultant to many National and International Institutions such as the International Institute of Tropical Agriculture, Federal Ministry of Environment, and our own Consultancy, Research and Development Centre (CORDEC) in the areas of Cytogenetics and Molecular Genetics, Ecology and Biodiversity Conservation, Environmental Impact Assessment, Environmental Auditing, Revegetation and Phytoremediation, Afforestation and Land Reclamation. He has served in Federal Government constituted Panels for review of more than 10 EIAs covering power generation, steel rolling mills and both upstream and downstream oil sectors.

Awards and Honours

Professor Osuji was conferred with an Award of Excellence for his Contribution and Support to Nation Building and Youth Development in Nigeria in October 2007 by the National Association of Plant Science and Biotechnology Students. He received, in March this year, the coveted Paul Harris Fellow Award by Rotary International.

Private Life

Professor Osuji is a devoted Christian of the Catholic faith. He is presently married to Genevive Chimezie Osuji and their marriage is blessed with two daughters (Akunna and Kem) and a son (Obieze) all of whom are presently here. Our Inaugural Lecturer loves soccer, swimming and music.

Conclusion

Mr Vice-Chancellor, distinguished Ladies and Gentlemen, may I present an erudite scholar, a seasoned administrator, a devoted family man and Paul Harris Fellow of Rotary International as the 88th Inaugural Lecturer of the University of Port Harcourt, Professor Julian O. Osuji. Thank you.

Professor Hakeem B. Fawehinmi

28th June, 2012