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The Application of Genetics and Molecular Biology to Industrial Microorganisms Particularly Bacteria

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Genetics and molecular biology are important to the development of industries that employ biological processes. There are two dinstinct industrial uses of these tools: (1) increase in the production of existing biological substances and (2) development of microbial systems for the production of biological substances of animal and plant origins. In addition, the qualities of these products can be improved.

Key words: Genetics, Molecular biology, Industrial microbes, Biological processess.

Genetics

Genetics which is one of the most exciting fields of biological sicences is the discipline that deals with genes, characteristics of living things and the mechanisms by which the traits are passed from one organism to another (Madigan *et al*., 2001). The origin and development of this science started during the first third of the 19th century. However, Franscisco and John (1980), reported that the study of genetics began by an Austrian Monk, Gregor Mendel in 1856. He said that the Ausrian was the first to start genetical research carrying out many experiments on plants to study how hereditary characters are transmitted from parents to offsprings. These experiments resulted in a considerable body of knowledge concerning what traits are inherited and how these different expressions are related to each other for many organisms including fruit fly (*Drosophila melanogaster*), laboratory mouse (*Mus musculus*), corn (*Zea mays*) and tomato (*Lycopersicum esculentum*) (Burns, 1983).

The science of genetics has important practical applications. Based on Mendel's principle of selective breeding, it has been of great economic benefit to many countries (Burns, 1983). Plants and animals with desirable traits can be selected and cross-bred to produce new breed that has combined superior qualities of both the parental strains (Sarojini *et al*., 1984). In agriculture, cereal crops like wheat, corn, rice and others like groundnut, soybeans, sugar cane and potatoes have been selectively cultivated to improve their yields by increasing their resistance to diseases and other biological agents (Burns, 1983; Madigan *et al.,* 2001). In animal husbandry, animals of economic importance such as cattle, pigs, goats and poultry

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are constantly cross-bred to improve their performance. The use of animals in biological research has led to many discoveries. Thomas Morgan and his workers have contributed greatly to the study of genetics using animals as models. They were the first to use the common fruit fly (*Drosophila melanogaster*) for genetic studies by mapping out the chromosomes at the exact positions of the genes on each chromosome (Burns, 1983).

Genetics is a major research tool in the understanding of the molecular mechanisms by which cells function. It provides the approaches to the introduction of new properties into organisms (Madigan *et al*., 2001; Tortora *et al*., 2002; Taggart and Starr, 2006). Thus, genetics has many industrial applications. The most significant application of genetics centers on the fact that the basic unit component of all life is the cell. A cell is a physical compartment in which the complex biochemical reactions of living organisms occur (Crafts-Lighty, 1986). Each cell contains genes that comprise stretches of DNA; a polymeric chemical substance, which specifies the genetic heritage of that cell. It codes for protein molecules. The DNA is a long molecule made up of four different types of similar compounds called nucleotides. These compounds are made up of 4 "bases" named Adenine, Thymine, Cytosine and Guanine. In order to pass genetic information to their offsprings or generations and replicate themselves, cells divide. A DNA however, does more than just to replicate itself, but synthesize various biological substances through different complex biochemical processes using the sequence of the four bases. Ribonucleic acid (RNA) is a chain of nucleotides.

Proteins are another type of biological polymer but they are built up out of amino acids. The amino acids are about 20 different common naturally occurring kinds. Proteins are very important because they provide most of the structural components and many can catalyse chemical reactions. The basic process by which DNA makes RNA and then protein (gene expression) was mainly elucidated during the late 1960s (Crafts-Lighty, 1986). He said that the existence of inherited genes for specific traits was demonstrated in the 19th century but the nature of the genetic material was then unknown. Today, completely new discipline 'Biotechnology' which deals with all phases of the production of useful substances has emerged using well characterized genetic constituents. Biotechnology is underutilized in underdeveloped or developing countries because the materials and equipments required are very expensive. In developed countries like USA and Britain, these tools have been widely harnessed to improve industrial processes and establish new industries which generate revenue for them. In this review, we discuss the genetic tools and molecular biology techniques applied to manipulate indusrtrially relavant microbes with emphasis on bacteria, to effect the abundant manufacture of good quality products.

Microbial Genetics

An industry's primary aim is to maximize profit and there is the need for such industry to improve the strains of the microorganisms or to keep on manipulating the genes of the microbes involved in any production processes for better yields in terms of quality and quantity. To appreciate the process of strain improvement, adequate consideration should be given to the ability of an organism to make a particular product which is predicted by the genetic make-up of the organism (Okafor, 1987; Prescott *et al*., 2002).

Manipulation of the genomes of industrially important microbes

The methods employed for the genetic manipulation of the industrially important microorganisms are grouped into two major classes. These are: (1) methods which do not involve foreign DNA (mutations and gene amplification) and (2) methods involving foreign DNA (Recombination). Transduction, transformation, conjugation and genetic engineering use foreign DNA. The first class of methods particularly mutation has been extensively used to manipulate genes while the second one which is commonly end up in genetic engineering is a new tool with vast potentials for the diversification of products (Okafor, 1987; Maki, 2002; Prescott *et al*., 2002).

Mutations

Any change in the sequence of the four nucleic acid bases in DNA is known as mutation and this leads to a change in the properties of the organism (Okafor, 1987; Prescott *et al*., 2002).

Mutation can be spontaneous or induced by mutagens (or mutagenic agents) which can either be physical or chemical. Spontaneous mutation occurs rarely at low frequency of 10-6 to 10-9. Various agents used to induce mutation are of two main types viz: physical and chemical agents.

Physical agents

Ionizing radiations

These agents cause ionization in the molecules of DNA leading to the production of a highly reactive radicals such as peroxide and hydroxyl ions which cause oxidation and eventual breakage of the DNA strand. The disadvantages of using this method are that the radiation equipments are expensive and radiation is apt to cause breakage in the chromosomes (Hopwood, 1970; Brock *et al*., 1984; Tortora *et al*., 2002).

Ultraviolet rays

The microbes are treated with UV. Bacteria are exposed to UV wavelengths between 200-300 nm for varying periods lasting from 30 seconds to 20 minutes depending on the sensititvity of the organisms. The UV causes dimerization between pyrimidine residues in DNA thus inhibiting DNA replication (Tortora *et al*., 2002). (iii) **Heat:** High temperatures are used to mutagenize microbial cells. Heat brings about deamination of cytosine to form uracil as nitrous acid. This causes denaturation of DNA strands with point or deletion mutations (Brown, 1992).

Chemical mutagens

Chemicals act on non-dividing cells. Commonly used chemicals include deaminating compounds such as nitrous acid and hydroxylamine and alkylating agents (nitrogen mustard, nitrosoguanidine (NTG) or N-methyl-N-nitro-guanidine (MNNG) and ethylethane sulphonate (EES) or ethylmethane sulphonate (EMS)). Alkylating agents cause transition and transversion of bases, interstrand linkage of purine bases and deletions. The deaminating agents deaminate nucleotides causing oxidative deamination, transition and mispairing (Maki, 2002). Other chemicals are base analogues (2-Aminopurine and 5-Bromouracil) and intercalating agents (Acridine and Ethidium bromide). The base analogues which resemble the bases structurally replace the nucleotides resulting in transitions causing point mutation. The intercalating substances insert themselves between DNA bases causing addition or deletion of bases resulting in frameshift mutation (Prescott *et al*., 2002).

Gene amplification

Genes are amplified by increasing the number of copies of the existing genes so that the cells will make more of the product encoded by the genes. For example, if a gene is amplified so that 1,000 copies of the gene exists, then the cells will make 1,000 times more of the corresponding product. This method has good industrial application (Okafor, 1987). This is because a line of genetically identical organisms can be propagated and grown in bulk.

Transduction

This is the transfer of bacterial DNA from one bacterial cell to another by the use of a phage. The phage attaches to and lyses the cell wall of its host. It injects its DNA (or RNA) into the host. The viral genome then directs the host DNA to produce many copies of the phage, thereby increasing the number of the gene. This has good industrial potential (Okafor, 1987; Taggart and Starr, 2006).

Transformation

This is a change in the genetic property of a bacterium which is brought about when foreign DNA is absorbed by, and integrates into the genome of the donor cell. Transformation of many microorganisms has been successfully made. An example of *Bacillus* species showed that an inactive strain of *Bacillus* was transformed to one producing an antibiotic, bacitracin. This method has also been used to increase the level of protease amylase production in *Bacillus* species. The method therefore has good industrial potential (Okafor, 1987; Maki, 2002).

Conjugation

This involves transfer of plasmids or DNA between microbes by cell to cell contact or through pili (Okafor, 1987). In conjugation, the donor cell transmits genetic information to another cell, the recipient (Brock *et al*., 1984; Taggart and Starr, 2006).

Genetic engineering

This process is also referred to as Genetic Manipulation or Molecular Cloning or Gene Cloning. In genetic engineering, a cell is altered so that it can produce more or different chemicals or perform better or carry out new functions. The aspect of genetic engineering in microbial genetics that has become most widely known is Recombinant DNA (rec. DNA) Technology. This involves a group of techniques which allow pieces of DNA from a plant, animal or microorganism to be transferred to a host microorganism. By this technology, DNA is incorporated into the genome of the microbe thereby acquiring new abilities for the synthesis of substances or other biochemical transformations (Jacobsson *et al*., 1986; Taggart and Starr, 2006).

Most of the basic scientific works in this area are carried out using the bacterium, *Escherichia coli* as the host organism; although it is now possible to use a variety of other microorgansims as hosts. The technical tool used for the transfer of genetic information from a donor organism into a host organism include vectors such as bactriophages and restriction enzymes. Restriction enzymes are made by bacteria and they cut DNA molecules at places where there is a specific sequence of bases. The vectors are capable of moving from organism to organism and of reproducing themselves as the cells divide. The restriction endonucleases allow the cut of the donor DNA molecule into segments and insert desired piece into a vector. This vector then carries the donor DNA into the host (Fig 1).

Application and importance of bacterial genetics in industries

Agriculture

One of the most practical applications of microbial genetics to agriculture is in biological control. In biological control of insects, natural *Bacillus thuringiensis* was used to control gypsy moth. The insect-killing toxin genes of the bacterium, often found in diseased insects or in soil or plant debris, were transferred to some plants to protect them against insect attack. The insect's resistant genes were successfully transferred into tomatoes, tobacco and cotton. The genes produce the toxins in the form of crystalline proteins. A report shown in a Mansanto experiment, that tomatoes with this organsm's gene were completely protected from an attack by caterpillars that stripped other unprotected plants in the same field down to their stalks. Tobacco is protected from the tobacco horn worm with the same gene. This technique has also proved successful with rice. Resistant rice variety has been developed using transferred genes from *B. thuringiensis*. Other researchers have worked on bacterial strains

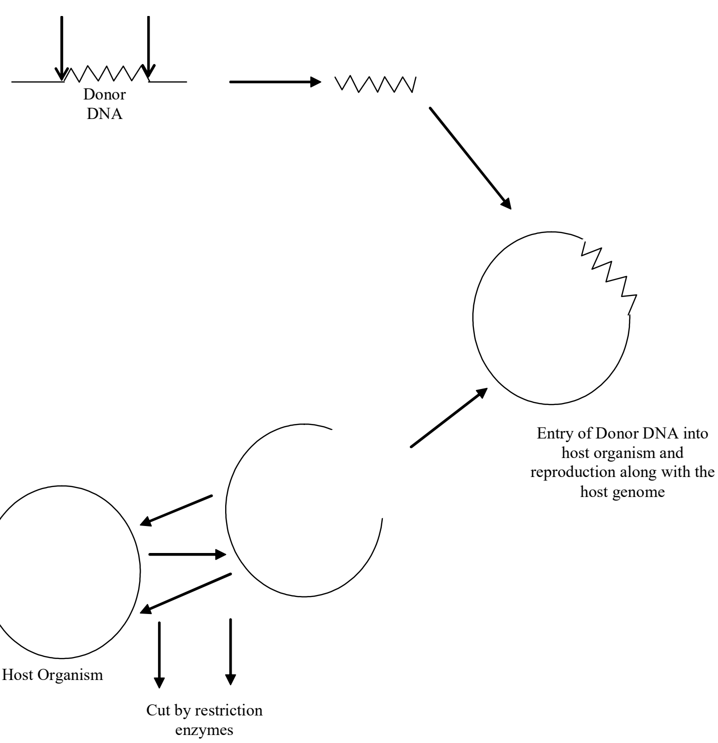

Fig. 1. Movement of DNA from Organism to Organism (Jacobsson *et al*., 1986)

carrying genes that kill rice pests such as brown plant-hopper, a vector of rice tungro virus (Robert, 1990). These plants offered the farmers many advantages of labour-free, protection of every part of the plant in every season, parts difficult to reach with sprays are protected from crop eating insects and the confinement of the pesticides to the plants, so leaving soil and ground unaffected. In addition, some newly developed strains of *B. sphararicus* have proved very effective against mosquitoes in trials around Mandras and Madurai and have remained active for some days (Robert, 1990).

Genetically improved bacteria are used to prevent frost damage which is a damage done to strawberry in Northern Asia. In an experiment carried out by Steven Lindow (cited by Sylvester and Klotz, 1983) involving the replacement of "ice-plus" bacteria with "ice-minus" types, a plant was protected from wild frosts. In addition, in *Rhizobium*-legume, improved strains and varieties of micro- and macro-symbionts are developed using molecular genetic techniques for farmers use to boost agriculture. Boboye *et al*. (2008c submitted) identified a 32 kb insert DNA from the genomic library of *R. fredii* USDA257 which extend host spectrum of a broad host range, *R*. species NGR234 to fix nitrogen on *Glycine max* cultivar Peking. Similarly, using molecular biology tool, Boboye *et al*. (2008b) sequenced the insert DNA to show the various genes borne on the fragment. Also, in plant-microbe interaction, the control of the development of symbiotic system of pea using its genetic system is found useful in sustainable agriculture (Borisov *et al*., 2006). Success of nitrogen fixation in this field will lead to less use of chemical nitrogenous fertilizers, and consequently reduce the energy used in producing them, eliminate the problems of pollution associated with their use and direct the raw materials to other productive activities (Okafor, 1987). Table 1 shows some genetically modified bacteria used in agriculture

Pollution control industry

Pollution of the environment is a major concern to the populace because of its detrimental effects on plants and animals both directly and indirectly (Ogiri, 2001; Agbogidi, 2003). Bioremediation (The use of microorganisms to return the environment altered by contaminants to its original condition (Okon and Trego-Hernandey, 2006)), is the most promising recovery method (Singh *et al*., 2001). Application of molecular genetics to bioremediation has greatly improved the capability of indigenous microbes to biograde

Table 1. Some genetically improved bacteria used in Agriculture

S.No.	Genes	Source of genes	Potential signifcance
1.	Gene Transfer vector	*Agrobacterium tumefaciens*	Insertion of new genes into crop plants for pest resistance.
2.	Nitrogen fixation ("nif" genes)	*Klebsiella* / *Rhizobium*	New strains of nitrogen fixing bacteria for increased legume yield.
3.	Nitrogen fixation ("hip" genes)	*Alcaligenes* /*Rhizobium*	Enhance energy efficiency of symbiotic nitrogen fixation in legumes for improved yields.
4.	Denitrification ("Den")	*Klebsiella*/*Pseudomonas*	Important in soil fertility and water quality.
5.	Physiological strain ("OSM")	*E.coli*/ *Salmonella*	Osmoregulatory gene provide tolerance to drought, salt and thermal stresses in microbes and plants.
6.	Photosynthesis "cfx"	Alcaligenes	Enhance efficiency of photosyntheticcarbon dioxide fixation in plants.
7.	Insect-killing toxin-gene	*Bacillus thuringiensis*	Pest resistance.
8.	"Ice minus gene"	*Pseudomonas*	Protection from frost damage.

Adapted from: Morris *et al*. (1983); Prescott *et al*. (2002); Taggart and Starr (2006).

pollutants. The first bacterium to be patented by a Canadian Biologist A. M. Chakrabarty and his colleagues at the University of Illinuos have engineered the development of bacteria that degrade 2,4,5,-T, an ubiquitous persistent and highly toxic herbicide commonly known as Agent Orange (Morris, 1983). The scientists co-cultured the microbial strain isolated from herbicide-contaminated soils with strains of *Pseudomonas* known to contain plasmids with biodegradative acitivities. The mixed microbial culture was adapted over a period of 8-10 months to grow in the presence of increasing concentration of 2,4,5-T. Heavily conrtaminated areas, such as those laid to waste over 15 years ago by U.S. Airforce target practice with Agent Orange can now be cleared up in a matter of weeks (Morris, 1983: Old and Primrose, 1986). Scientists at the Batelle Memorial Institute in Columbus, Ohio were engaged in genetic engineering of microbes that efficiently degrade chlorinated herbicides 2,4,-D and atrazine.

Furthermore, the use of microorganisms (Biopesticides) for environmental control of mosquito has been successful in eradication of insect borne diseases. In the Malaria parasite biocontrol, genetical development of a bacterium (*Bacillus thuringiensis* serovar israeliensis (BTI)) and *B. sphaericus* has been achieved. Biopesticides are now being used in worldwide field test designed to control the population of mosquitoes (Phillip, 2001; Shilulu *et al*., 2002). Although, search for better strain to meet with the need of certain countries continues by the use of molecular genetic. This is because biopesticides compare considerably with conventional chemical pesticides in efficacy and cost (Keya and Lacey, 2007). Recently, *Rhizobium*-legume symbiosis was found to be useful to rhizoremediate heavy metals (Pajuelo *et al*., 2006). In a bid to intensify the use of genetically modified microbes to control environmental pollution over two decades ago, SRL International undertook a program to compile a list of common toxic chemicals that are amenable to microbial biodegradation and to isolate an engineered improved strains that might have commercial value (Morris, 1983). Some genetically improved bacteria used in pollution control are presented in table 2.

Mining industry

Mining involves recovery of metals from their ores and petroleum from oil shales. The recent use of microbes in mining industry employs genetic manipulation of microbes (Biotechnology). Microbes such as *Thiobacillus ferroxidans* is considered to play a major role in most microbial leaching operations. In view of the potential benefits of recombinant DNA technology, *T. ferroxidans* was genetically manipulated to produce strains that have enhanced leaching capabilities. The development of genetic system for *T. ferroxidans* and an understanding of gene expression in acidophilic autotrophs are important areas that research has concentrated on in leaching. Thiobacilli are able to develop considerable resistance to the very high concentration of the

Table 2. Some genetically improved bacteria used in pollution

S.No.	Pollutants	Bacteria
1	Petroleum hydrocarbon	*Acinetobacter*, *Arthrobacter* *Mycobacterium* and *Pseudomonas*
2	Pesticide/herbicides e.g. aldrin, dieldrin. Organophosphorus type e.g. Parathion, Melathion. 2,4-D ketone and Piperonylic acid.	*Pseudomonas* and *Arthrobacter*.
3	Other chemicals Bis (2-ethylhexyl) phthalate. Dimethylnitrosoamine	*Serratia marcescens* Photosynthetic bacteria
4	Ligno cellulosic waste Municipal waste pulp, Pulp mill, Lignins, Phenols.	*Pseudomonas*, *Thermospira* *Arthrobacter*, *Chromobacter* *Xanthomonas*.

Source: Morris (1983); Tortora *et al*. (2002).

metals being leached, but the bacterium is inhibited by some metals such as silver, mercury and cadium at quite low concentrations. In order to obtain *T. ferroxidans* that is resistant to these elements, Morris (1983) stated the possibility of isolating appropriate plasmids from other bacteria and introducing them into Thiohacilli using recombinant DNA.

Thiobacillus thiooxidans and *T. ferroxidans* are used to recover uranium from low grade ores, through bioleaching by solubilising the metal (Sharma, 2005). Bioleaching also occurs with fungi. *Aspergillus niger* and *Penicillium simplicissimum* are able to solubilize copper and tin by 65%, and aluminium, nickel, lead and zinc by more than 95% (Brandl, 2001). *Xanthomonas campestris* and thiobacilli are used in the recovery of petroleum. The former is employed in the tertiary recovery of petroleum (Sharma, 2005). Improved bacterial growth and mineral leaching activity was obtained when Thiobacilli were grown in conjunction with the nitrogen fixing bacterium, *Beijerinckia lacticogenes* which supplied the bacteria with nitrogenous nutrients (Morris, 1983). Morris later stated that, it is worthwhile introducing the nitrogen fixation genes (*nif* genes) directly into the *Thiobacillus* and that *nif* genes from *Azotobacter* or *Klebsiella* could be utilized since they share similar structural and biochemical features with *Thiobacillus*. Other genetically improved bacteria used in mining industry are *Thiobacillus ferroxidans*, *Leptospirillum ferroxidans, Sulfolobus* species, Thermophilic thiobacilli, Thermophilic *Thermosulfidooxidans* species (Bull *et al*., 1979).

Pharmaceutical indutry

There are many pharmaceutical products obtained from plants and animals. Many of them are of animal origin. Interferon, a protein synthesized by most cells of higher organisms in response to viral infections is produced by applying bacterial genetics. Interest has grown in the production of interferons because of their anti-viral and anti-tumor potentials (Brock *et al*., 1984). Human interferon was earlier produced from cell cultures of leukocytes, lymphoblasts, or fibroblasts. This method is tedious, very expensive and its yields are extremely low. Recently, interferon has been produced more cheaply by genetic engineering. Although the 'interferon' so produced lacked the carbohydrate moeity found in the animal type, it was just as active as the human type when tested for its anti-viral activity in monkey. Diabetes is currently treated with insulin obtained from animals. Not only is insulin expensive, but also some patients react to animal insulin. Microbial insulin is not only cheaper than animal type but it does not contain body components of cattle and pigs which cause allergy in some patients. Cloning of mammalian insulin gene in *E. coli* has been successfully done (Brock *et al*., 1984; Madigan *et al*., 2001).

A number of mammalian proteins are of great medical and commercial interests. Commercial production of human proteins by direct isolation from tissues or fluids is complicated and expensive or even impossible; cloning the gene into bacteria from a human protein makes the commercial production possible. Table 3 provides a list of some mammalian genes which have been expressed in bacteria and their commercial or medical importance.

Frequently, killed virus preparation are used as vaccines which are materials that can induce immunity to an infectious agent. The limitation experienced in the use of this kind of vaccine is that there is always a potential danger to the patient, if the virus has not yet been completely inactivated. The active ingredient in the killed virus vaccine is the protein coat and by genetic engineering, the viral coat protein genes have been cloned and expressed in the bacterium *E. coli* (Fig. 2) making possible the development of safe and convenient vaccines (Brock *et al*., 1984).

The development of resistance to existing antibiotics by pathogenic microorganisms have called for search for new antimicrobial agents from plant extracts against pathogenic microorganisms. This requires the application of genetics and molecular biology. Recently many research works were done on plant extracts for use against pathogenic microbes. These include a previous work done by Boboye *et al*. (2007) who noted that *Capsicum annum* and *C. frutescens* inhibited the growth of *Klebsiella pneumoniae*, *Streptococcus faecalis*, *Corynebacterium diphtheriae*, *Pseudomonas aeruginosa* and *Escherichia coli*. Other scientists reported similarly that *Capsicum* *annum* possesses antibacterial, antifungal and antiviral activities been active against both gram positive and gram negative bacteria such as *Staphylococcus aureus*, *Listeria monocytogenes*, *Bacillus subtilis*, *Escherichia coli*, *Pseudomonas aureginosa*, fungi like *Candida tropicalis*, *Saccharomyces cerevisiae* ATCC 9763 and Herpes zooster virus (Kivanc and Akgul, 1998; Barber *et al*., 2000; Dorantes *et al*., 2000; Cereaga *et al*., 2003; Farag *et al*., 2003). To study the basis underlying a particular reaction of microbes to plant extracts, Boboye and Odekunle (2008) showed that EMS regulated sensitivity of *Staphylococcus aureus* to sweet pepper (*Capsicum annum*). The use of transpsons to manipulate antibiotic related genes in bacteria to elicit over-production of antibiotic intermediates is also being considered in genetic manipulation. Transpsons are genetic element that facilitate the movement of adjacent genes from one site on the DNA to another (Madigan *et al*., 2001).

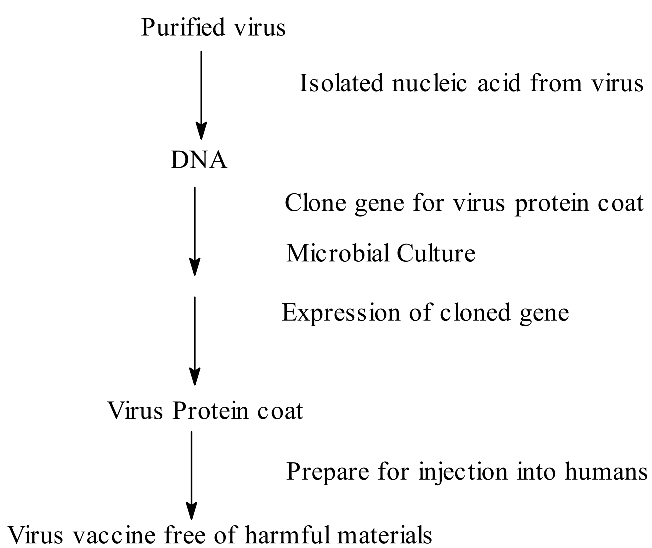

Fig. 2. Steps used in the preparation of a virus vaccine by genetic engineering (Brock *et al.*, 1984; Madigan *et al.*, 2001)

Table 3. Mammalian genes expressed in *Escherichia coli*

S. No.	Protein	Function
1.	Interferon	Anti-viral agent, anticancer
2.	Insulin	Treatment of diabetes
3.	Serum albumin	Transfussion application
4.	Growth hormone	Growth defect
5.	Urokinase	Blood clotting disorders
6.	Parathroid hormone	Calcium regulation
7.	Human virus (Hepatitis B Cytomegalovirus influenza)	Vaccines
8.	Animal viruses (Foot and Mouth disease)	Vaccines

Source: Madigan *et al*. (2001).

Chemical and food industries

The potential applications of genetic engineering in the chemical industry lies largely in the area of producing organic compunds. In 1988, the United States of America succeeded in cloning a gene encoding a polymer making use of genes of a species of *Alcaligenes* (Robert, 1990). This bacterium swell to make 80% of their dry weight in the form of a biopolymer called polyhydroxybutyrate (PHB) which acts as an energy storage substance when they are threatened by a reduction in their nitrogen supply. By continous cultivation of a mutant strain of *Klebsiella aerogenes* in xylitol (a sugar rarely found in nature), a strain with greatly amplified enzyme system for utilizing the sugar was developed (Okafor, 1987). A Japanese firm, Ajinomoto patented a recombinant strain of *E. coli* that contains multiple copies of lysine-encoding gene borne on plasmids (Morris, 1983).

Ethanol production by thermophilic bacteria, *Clostridium thermocellum* and *Thermoanaerobium brockii* was enhanced by obtaining mutant strains lacking lactate dehydrogenase (Phelps and Clarke, 1983). Moris (1983) also reported that genes have been cloned in several laboratories to increase the level of alcohol dehydrogenase in microbial cells using genetic engineering technique. Ethanol obtained this way, can be used in the manufacture of perfumes, flavoring extracts, high quality medicines, organic solvents and preservatives. Genetic engineering has played a significant role in the development of biogas production. It is possible through this technique to combine the acidogenic and methanogenic activities in a single microbial population so that the proper balance of activities is maintained irrespective of population size. Table 4 shows some bacteria genetically modified to synthesize chemicals. The application of genetic and molecular biology to the synthesis of various substances in food industry has been extensively researched into. Enzyme production in bacteria can be increased up to 100-fold by gene amplification using phage or plasmids. Various enzymes, amino acids and vitamins in current use were obtained from this development (Okafor, 1987). Examples of this amplification was observed in long-term chemostat studies with *E. coli* in which strains capable of producing extremely high levels of β-galactosidase (enzyme catalysing the utilization of lactose) were produced made, limiting amounts of lactose. In this example, the enzyme formed was 25% of the total protein produced by the organism (Okafor, 1987). The synthesis of pectate lyase in *Xanthomonas campestris* was mutagenically controlled to obtain various mutants among which are super-inducble variants that showed higher level of enzyme induction activity than the parental strain of the bacterium (Boboye and Shonukan, 1993). Also, Boboye and Alao (2008) in an attempt to obtain a strain of a tropical *Rhizobium* species F1 that could produce trehalose-catabolic-enzyme more than the wild-type bacterium, they chemically mutated the bacterium. The result of their work showed that a class of the mutants (super-trehalose-catabolic-enzyme producers) were superior to the parental strain. These super-trehalose-catabolic-enzyme producers are useful to detoxify trehalose; the sugar is toxic to certain plants (Veluthambi *et al.*,

Table 4. Some genetically improved bacteria used in chemical industry

S. No.	Microbes	Products
1.	*Alcaligens* species	Polyhydroxybutyrate
2.	*E. coli*	β-galactosidase
3.	*Klebsiella aerogenes*	Xylitol
4.	*Bacillus*	Bacitracin
5.	*E. coli*	Lysine
5.	*Clostridium thermocellum*	Ethanol
6.	*Thermoanaerobium brockii*	Ethanol

Adapted from Morris (1983); Okafor (1987) and Madigan *et al*. (2001).

1981). This is critical in plant-microbe interaction when rhizobia-legume symbiosis is considered.

In brewing and baking industries, high ethanol and carbon dioxide synthesizing strains of yeasts have been developed genetically. More research is carried out to further improve these strains or search for naturally genetically manipulated yeasts. A work done by Boboye and Oigiangbe (2008) on the effect of EMS on sucrose-degrading-enzyme of *Candida versatalis* strain 'Bol 1' showed that a variant of the yeast could degrade sucrose (a sugar commonly used in baking) more than the wild-type strain. This manipulation enhanced the release of more simple sugar necessary for the synthesis of increased amount of CO_2 during fermentation of the baking process. Also, to improve baking, an amylase was cloned in *Saccharomyces cerevisiae* by Xiaola *et al*. (2002). Various reports are available on the improvement of yeast for single cell protein. This is related to ability of the yeast grow to fast and produce essential nutrients (amino acids and vitamins).

CONCLUSION

Applications of genetics to bacteria important in various industries have many advantages as described above. In agriculture, useful plants are being protected from insect pests and various biological damages therefore, preventing or reducing food shortages from genetic. Biological control of these bacteria used in agriculture offers greater advantages over chemical control. In pollution industry, microbial degradation of various herbicides, pesticides, petroleum hydrocarbon and other chemicals which are very toxic and can cause serious pollution in the environment are curbed. Improved mineral leaching activity are achieved using recombinant DNA with the bacteria such as Thiobacilli. Also, bacteria genetic has proved useful in medical line. Products which improve the supply of drugs are increased to reduce enormous unaffordable cost and enhance the quality of the products. Thus, these industries are able to achieve their primary aims of maximizing profit and manufacturing products of high quality in terms of physical qualities such as flavour, colour, texture, aroma, purity and product safety.

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