

## Research Article

# The Impact of Sequencing Human Genome on Climate Change & A Novel Approach to Reducing CO<sub>2</sub> By Spreading Chloroplast Genome

**Hameed Khan**

Senior Scientist, Department of Genetics & Robotics, NCMRR (National Center for Medical Rehabilitation Research), National Institutes of Health (NIH), Bethesda, Maryland

**Abstract**

This abstract presents a novel approach to reduce the pollutant Carbon dioxide from the atmosphere by spreading the cloned Chloroplast genome in plant microbial life on the surface of oceans across the world. Photosynthesis in plants is carried out in Chloroplast genome which can absorb Carbon dioxide from the atmosphere in the presence of sunlight to conduct photosynthesis by converting Carbon dioxide to its food Carbohydrate and releasing Oxygen as its by-product. During sequencing Chloroplast Genome, we will splice essential amino acid genes in the Chloroplast Genome, it will not only remove CO<sub>2</sub> from the atmosphere, but also will produce Oxygen and essential amino acid rich carbohydrate compounds which could be used for human consumption. This abstract also describes how the Industrial Revolution increased the level of Carbon dioxide from 175 ppm pre-industrial level to the present 400 ppm and most likely to increase to 800 ppm by the end of this century. To prevent environmental collapse of our planet, we need to discover novel approaches to reduce all environmental pollutants especially Carbon dioxide. We need energy to run the engine of modern society, we need to discover alternate source of energy. Instead of fossil fuel (main source of Carbon dioxide), we must develop other sources of energy such as Wind Turbine, Geothermal, Solar Panel, Liquid Hydrogen and Fusion reaction. Using Genetic toolkit developed for sequencing Human Genome, we could sequence Chloroplast Genome for Genetic Engineering. We can teach the next generation of scientists how to cut, paste and copy Chloroplast genome enrich with essential amino acids Codons in plant microbial life form for worldwide release. May be within a hundred year, we might be able to bring back the level of Carbon dioxide to a more acceptable level.

**Keywords:** Chloroplast, Photosynthesis, Human Genome, Genetic Toolkit, Codons.

**Historical Background**

We have polluted the water; we have polluted the air and we have polluted the land. Today, we wonder if the water we drink is safe; the air we breathe is safe; and the food we eat is safe. The world population of over seven and a half billion people emits about 120 million ton of Carbon dioxide each year in the atmosphere. Increased level of Carbon dioxide is responsible for Climate Change. Sunlight coming from Sun strike Earth surface and return to atmosphere as infrared light. Pollutant such as Carbon dioxide, Methane and Nitric oxide act as a Glass Ceiling of a Green House which traps the heat energy and increase the temperature worldwide causing Greenhouse Effect. Carbon dioxide is generated by burning fossil fuel such as coal, oil and natural gas in power plants and automobile. Beside Carbon dioxide methane is another major contributor of the Greenhouse Gas produced by farm animals by digesting starch producing Methane and Nitric oxide which is also produced by automobiles combustion in their engine by burning oil.

What concerns us most is the level of Carbon dioxide which has gone up from the pre-industrial level of 175 ppm to 400 ppm today. If we continue to pump the Carbon dioxide in the atmosphere with the current rate, by 2050, the level Carbon dioxide would likely to be 800 ppm. When most people of my generation would be dead and gone, you and your children would confront the reality. To get the evidence, scientists conduct experiment. Trust the results of bench scientists because their work is peer-reviewed by experts in the field before publications. For selfish reason, we cannot conduct the experiment not even once to determine the threshold level of Carbon dioxide on Earth beyond which the Greenhouse Effect cannot be reversed. The best evidence is obtained by looking at the neighboring planets. Two sister planets of Earth, Venus and Mars are almost equal in size and yet have such a different climate. Planet Venus is filled with Carbon dioxide, and it has a temperature of 900-degree Fahrenheit; hot enough to melt metal lead while Mars has

**\*Corresponding Author:**

Hameed Khan, Senior Scientist, Department of Genetics & Robotics, NCMRR (National Center for Medical Rehabilitation Research), National Institutes of Health (NIH), Bethesda, Maryland;  
E-Mail: [Hameedkhan111@Comcast.net](mailto:Hameedkhan111@Comcast.net)

**Received Date:** 25 Sep, 2021

**Accepted Date:** 30 Sep, 2021

**Published Date:** 04 Oct, 2021

a zero degree temperature. No life form could survive in such harsh conditions. Although Venus is closer to Sun, the hot temperature is not due to closeness to the Sun, but because of the Greenhouse Effect. The cloud of Venus is made of Carbon dioxide. We expect that the cloud of Venus should reflect the sunlight and cool the planet instead the high level of Carbon dioxide traps the Sun's energy and heat up the internal environment raising the high temperature. If we continue to pump Carbon dioxide in our atmosphere with the current rate, may be within a hundred-year, Earth's climate will be on its way to become like the climate on Venus. Most scientists believe that the summer of 1988 was the first alert for us all.

On the other hand, planet Mars has no atmosphere, no Oxygen, no Ozone, and no Carbon dioxide to hold Sun's heat to warm the planet. The entire planet is extremely cold and is like a big Ozone Hole allowing the Sun's Ultraviolet radiations to bombard the Mar's surface sterilizing the planet. Earth would have the same fate as Mars if we had not passed the Montreal Accord in 1989 banning the use of Freon gas in our cooling systems which destroys the Ozone shield protecting life on Earth. We need Montreal type Accord to reduce the level of Carbon dioxide to pre-industrial level that is around half the current level.

In this article, I will attempt to answer how can we maintain the balance between the Industrial development and the environmental pollution? We cannot take one and leave the other. We cannot live in today's comfortable life with the climate of Moses time. I have divided my presentation in three parts: first, I will describe the pollutants responsible for causing the climate change; second, I will describe how can we reduce the level of pollutants and finally what if we ignore the threat and go on living as if nothing will happen? Two important items you take from this article. First, think critically and second look for the evidence.

First, let me begin with the historical background. Savant Arrhenius in 1889 accurately measured the level of Carbon dioxide in the atmosphere to be 175 part per million (ppm). Because of the Industrial Revolution and its contribution, today, the level of Carbon dioxide has gone up to 400 ppm. As I said above, with the current rate of increment, by 2050, the Carbon dioxide level is expected go up to 800 ppm as the population will increase from 7 and a half billion to 9 billion, (we are adding about 90 million people each year to our planet). Even a few degree rise of temperature worldwide would have catastrophic effect by disrupting weather system causing extreme tornados, massive hurricanes, and uncontrollable cyclones. The warming will melt the polar ice and glaciers causing sea level rise. As the population increases so does the Industrial pollution and there is a global temperature rise and the expected increase of temperature worldwide by +1.2 degree centigrade by 2020.

Our Solar System was formed about four and a half billion years ago. Within the first billion years, atmosphere was cool enough to produce the first Sunshine, the first rainfall and the first lightening. A million-lightning strike planet Earth each day. The first organic molecules, the nucleotide, the building blocks of our life, appeared when the lightning struck at a phosphate rock containing a collection of gases including Methane, Carbon dioxide, Nitrogen in a water pond at some remote corner of the Earth. The intense heat generated by the lightening combine the gaseous mixture producing the first organic molecule the nucleotide RNA (Ribonucleic Acid).

(In 1953, Stanley Miller at Chicago University created the early environment by mixing the above atmospheric gases in a flask containing water and generated organic molecules by sparking electricity for a week. He created amino acids the first organic molecules serve as the building block of life.) The other experimenters created nucleotides which form RNA and DNA. The RNA molecule alone can create life. It can store information like DNA (Deoxyribonucleic Acid) and acts as

protein to catalyze the reaction. The RNA world has no Oxygen. The early life was anaerobic and could survive in the absence of Oxygen.

Soon after the Earth was formed about four and a half billion years ago, the surface of the Earth was like visions of hell filled with the oceans of liquid rock, boiling sulfur, and impact craters everywhere! Volcanoes blast off all over the place, and the rain of rocks and asteroids from space never ends. Within a billion year, the heavy bombardment slowed down, and the surface begins to cool. The heavy bombardment of comets which brought water to Earth. Every drop of water on Earth was brought by comets. Planet Earth was not only cooled but also submerged in Ice age. The climate of the late Precambrian time, the Pro-terozoic eon (2.5 billion years ago to 543 million years ago) was typically cold with glaciations spreading over much of the earth. At this time the continents were bunched up in to a single supercontinent. The first living creature appears within a billion year. Most of the life that existed during the Precambrian Time span were prokaryotic single-celled organisms. As the climate change over a billion year, there appeared the first sunshine, the first snow melts, the first rainfall and the appearance of first single cell Pre-Cambrian creatures that attack each other forming a multicellular creatures. The extinction of this life forms paved the way for a burst of new life, called the Cambrian explosion, during the following Paleozoic Era. These earliest forms of life resembled cyanobacteria. They were photosynthetic blue-green algae that thrived in the extremely hot, carbon dioxide-rich atmosphere. For millions of years, the job of blue green algae was to perform photosynthesis that is to absorb Carbon dioxide and release Oxygen.

Plants first appeared on the Planet Earth about 400 million years ago. With the appearance of plants, Oxygen molecules accumulate in abundance. The photosynthetic apparatus, the chloroplast, in plants started absorbing Carbon dioxide and started releasing Oxygen in the atmosphere. As forests thrive in the aerobic atmosphere, the evolution in many plants played significant role in converting the Carbon dioxide to Oxygen. Of all the plants, Maize is the winner. The fields of plants maize can efficiently assimilate and convert CO<sub>2</sub> to carbon products such as Carbohydrates during photosynthesis releasing Oxygen. As the Sun rays strike the forests on Earth surface, the Chloroplasts in the trees convert the Physical energy into Chemical energy by photosynthesis. Chloroplast genome absorbs Carbon dioxide from the atmosphere and in the presence of Sunlight and Water; it converts the pollutant Carbon dioxide into its food Carbohydrate and release Oxygen. Most of the Earth is powered by photosynthesis. It was plants that introduced Oxygen in Earth's atmosphere.

We need energy to run the engine of modern society and we all would like to generate maximum amount of energy by producing least amount of Carbon dioxide. The first source of energy is obtained by using Solar Power. The Solar energy reach out to Earth is over 1000 Tero-Watts (TW) in the form of Sunlight. Sunlight comes in and turns its solar energy into the chemical energy in plants by Photosynthesis. The entire Earth surface receives about 1000 (TW) of energy each day. Our civilization uses about 20 TW of energy each day to run the modern civilization. We have to figure out how to use the enormous unused solar energy to flourish our society. We can partner with nature. It is a challenge for the engineering skill of human race to use this untapped energy for the benefit of mankind. As I said above, more than seven billion people live on Earth today. We are adding 90 million new mouths to feed each year. With the current rate of growth, the population is likely to be nine billion by 2050. To face the upcoming crisis, we need to train an army of young intellectuals in genetic engineering and biology challenging them to convert this untapped physical energy into chemical energy.

To run the engine of the modern society, we need electricity. To generate electricity, our power plants are fired by fossil fuels such as Coal, Petroleum, Natural gas, and Wood. By burning Carbon fuel

in our cars or in our power plants, we generate Carbon dioxide and oxides of Nitrogen which act as glass ceiling of a Greenhouse, and which retain heat raising internal temperature. Compared to Nitrogen gas (about 80 percent in our atmosphere) and Oxygen gas (about 20 percent) in our atmosphere, the amount of Carbon dioxide (about 0.04 percent) is negligible. Unfortunately, it is not the level of Carbon dioxide that concerns us; it is its function which acts as a glass ceiling of a greenhouse. The energy release by burning the fossil fuels in cars, power plants and industrial complexes generate Carbon dioxide which spread as a thin sheet covering the entire planet which does not allow the heat to escape but trap the heat like a Greenhouse. As a result of the trapping of this heat energy, it heats up the internal atmosphere raising temperature worldwide. It is this trapped heat which concerns us. In a Greenhouse, we can open the windows and let this heat out. Unfortunately, we don't have such window in our planet.

Worldwide rise in temperature, could also release the trapped Methane from the permafrost and polar ice sheet frozen over millennia. Methane is another greenhouse gas which is trapped in the ice sheet since the dawn of our planetary origin. The rise in temperature decreases the ice sheet of the polar cap releasing Methane on one hand and causing the rise in sea level on the other hand. Our challenge in the next century is not only to slow down the release of Greenhouse gases but also to stop and reverse the trends achieving the pre-industrial level. To replace the Green house gases, we have to develop alternative source of energy such as Wind turbine, Geo thermal energy, and solar panels.

All nations should be encouraged to become energy independent by massive reforestation of their countries. Mountains rich nations should use the huge Wind turbines to generate their national energy need. The Wind turbines could also be installed in shallow waters around the world. Desert rich nations could use Solar panels to generate their national energy needs. Nations who shared a major part of the arable part of the planet should be encouraged to use Chloroplast technology to reduce Carbon dioxide level. Farmers should be encouraged to use Chloroplast rich plants (such as Maize) to produce food providing grains with excessive amount of Oxygen while removing Carbon dioxide and to reduce the level of Methane. To prevent the production of excessive amount of Methane in the atmosphere, we have to reduce the level of live stocks. For meat eating population, we could develop the tissue-culture technology to produce highly nutritive edible meat. To reduce the level of Nitric Oxide from the atmosphere, we must develop Hydrogen run automobile engines.

Another major problem of increasing the level of Carbon dioxide is the acidification of the oceans. As the level of Carbon dioxide in our atmosphere increases, Ocean acidification is likely to increase. It is estimated that about 25 percent of all Carbon dioxide is absorbed in the oceans which is converted to Carbonic acid increasing the acidity of the oceans. With the current rate of Carbon dioxide emission, by end of the 21st century the level of Carbon dioxide is expected to increase by 170 percent which in turn will increase ocean acidity several folds. Acid sensitive species will suffer mass extinction. Shelled creatures will find difficult to make shell in high acidic medium affecting our sea food supply. Coral Reefs support about 25 percent of marine life around the world. Ocean acidification poses a serious global threat to coral reef formation.

Second, I will describe how can we reduce the level of pollutants? Our major source of energy should be Solar Panels and Wind turbines. If the petroleum at the gas pump station remains cheap for about three dollars a gallon, the technology is unlikely to develop. Increasing tax on Petroleum products will generate revenue to build roads and bridges. Do we have to use fossil fuels to generate electricity? No, we could use nuclear energy to generate electricity. It is the electricity which runs the engine of modern society. Nuclear energy provides unlimited amount of Greenhouse gas free energy. Unfortunately, it

also produces nuclear waste which last for generations. Major source of energy in France is nuclear. Their nuclear waste dumping ground is not in France but in their old colony in Africa that is in French Guyana. The Hanford nuclear waste site in the State of Washington is the prime example of our past failure to store nuclear waste safely for long period. The storage tank was installed in 1976 and it is supposed to store for 20 years. It has been over 40 years and tanks started leaking causing havoc. The flowing leak joined the Columbia River contaminating the Salmon fish with radioactive particles.

Nuclear energy could be obtained by two sources either by Fission Reaction or by Fusion Reaction. Today, we have 61 commercially operating nuclear power plants with 99 nuclear reactors in 30 states in the United States. They are generating electricity and are all powered by Nuclear Fission Reactors. Nuclear fission reactors generate dozens of radioactive by-products as nuclear waste which has extremely long half life. Radioactive waste is stored in thousands of drums buried underground and their radioactivity will last for generations. Ask people in Nevada who have been accepting nuclear waste from around the world for years how much they love nuclear junk in their backyard. A massive earthquake will throw millions of barrels on the surface releasing radioactivity creating havoc.

The second source of electricity generated by Nuclear Fusion Reaction which is taking place on the surface of Sun is not yet developed. We don't have powerful enough technology to fuse Hydrogen atoms to generate Helium and release sub-atomic particles which come to us as sunlight energy. Preliminary work on Fusion technology is being done in the Plasma Physics Lab at the Princeton University. We don't have to create Sun on Earth. We could use Solar Panels to convert the Sunlight into electricity. It is the safest way to produce unlimited amount of electricity. The other sources to generate electricity as I said earlier are to use large wind turbines, large number of Solar Panels and geothermal energy. These sources of producing energy do not add Carbon dioxide to our atmosphere.

How could we reduce the level of Carbon dioxide without increasing pollutant gases to run the engine of modern society? In this article, I am proposing an alternative way to reduce the level of Carbon dioxide from the atmosphere by inserting and spreading Chloroplast genome in non-Chloroplast microbial life to reduce the level of Carbon dioxide. I believe that it is possible to re-engineer nature to switch on Chloroplast genes in all green plants, microbes, algae, planktons, and krill for world-wide distribution. After the completion of the Human Genome Project about eighteen years ago, we realized that there is another way to remove excessive amount of Carbon dioxide from the atmosphere that is by introducing the photosynthetic apparatus Chloroplast genome in microbial life forms. Our work on the Human Genome Project provided us with the genetic toolkit to cut, paste, to shut off a gene or to copy to insert or to delete a gene from the Genome of all living creatures to produce new food, new fuel, and new medicine to treat every disease known to mankind. Using the same toolkit, we could clean up our environmental pollution by introducing Chloroplast Genome in bacterial life forms.

Our entire genome, the book of our life, is written in four nucleotides and they are A, (Adenine) T (Thiamine), G (Guanine) and C (Cytosine). The chain of these nucleotides forms a double stranded string called the DNA (Deoxy Ribonucleic Acid). According to Francis Crick's Central Dogma, [1] double stranded DNA is transcribed into a single stranded RNA which is translated in the Ribosome into proteins. The discovery of the double helical structure of DNA explained how the information to create life is stored, replicate, evolved and passed on to the next generation. This discovery opened a New World Order of ideas and buried the old explanation of the magical mystical appearance of life on Earth.

The double stranded DNA explained that the essence of life is information, and the information is located on these four nucleotides. Every set of three nucleotide on the mRNA forms a Codon which codes for a specific amino acid. The four-letter text of nucleotides forms a three letter Codon which codes for an amino acid. There are 64 different combinations of Codons which codes for all 20 amino acids.

Sequencing human genome identifies the number and the order of nucleotides in which they are arranged. Less than two percent of our genome contains regulatory region, a piece of DNA, which controls the function of genes. More than 300 regulatory regions have been identified. More than ninety eight percent of our Genome contains non-coding region used to be called Junk DNA which makes up to sixty percent of our entire Genome. The non-coding regions contains repetitive piece of DNA which is tightly packed and mostly remain silent. The sequencing of this region showed that the non-coding region is the part of Viruses and Bacteria picked up by our Genome during the million years of our evolutionary process. During Bacterial or Viral infection, the non-coding DNA could unfold transcribing into RNA resulting into hazardous protein which could create havoc for our health.

In 1990, US congress authorized \$3 billion to decipher the entire human genome within 15 years. The US Congress told us to complete the work by 2005 or all funding will be stopped. There were critiques who said reducing the mysteries of life to few organic molecules will not only violate the sanctity of life, it will also violate the laws of God and it will be sin. We will be in deep troubled in the afterlife. They asked us how you could decipher a human being made of one hundred trillion cells and each cell inheriting a string of three billion and two hundred million genetic letters from Mother and another set of three billion two hundred million from your father. Could this all be done in 15 years; no they said it could not be done in 15 years nor could it be done in 1500 years nor could be done in our lifetime on the face of the Earth?

We proved them wrong. An army of young intellectuals from six industrialized nations and twenty Biomedical Center joined our effort. This effort was led by US followed by Germany, France, England China and Japan. Within 13 years, the entire book of life was deciphered. The entire sequence of human genome was published in the Scientific Journal Nature and was linked to the website for the world to see. If you have an excess to a computer keyboard, you have excess to all that information.

We deciphered the entire Human Genome to map, identify and locate all genes under the title, "The Human Genome Project." We found that our genome contains six billion four hundred million nucleotides bases half comes from our father and another half comes from our mother. Less than two percent of our Genome contains genes which code for proteins. The other 98 percent of our genome contains switches, promoters, terminators etc. The 46 Chromosomes present in each cell of our body are the greatest library of the Human Book of Life on planet Earth. The Chromosomes carry genes which are written in nucleotides. Before sequencing (determining the number and the order of the four nucleotides on a Chromosomes), it is essential to know how many genes are present on each Chromosome in our Genome. The Human Genome Project has identified not only the number of nucleotides on each Chromosome, but also the number of genes on each chromosome.

The following list provide the details composition of human genome. It also provides the number of nucleotides, and the number of genes present on each Chromosome:

We found that the Chromosome-1 is the largest Chromosome carrying 263 million A, T, G and C nucleotides bases and it has only 2,610 genes. The Chromosome-2 contains 255 million nucleotides bases and has

only 1,748 genes. The Chromosome-3 contains 214 million nucleotide bases and carries 1,381 genes. The Chromosome-4 contains 203 million nucleotide bases and carries 1,024 genes. The Chromosome-5 contains 194 million nucleotide bases and carries 1,190 genes. The Chromosome-6 contains 183 million nucleotide bases and carries 1,394 genes. The Chromosome-7 contains 171 million nucleotide bases and carries 1,378 genes. The Chromosome-8 contains 155 million nucleotide bases and carries 927 genes. The Chromosome-9 contains 145 million nucleotide bases and carries 1,076 genes. The Chromosome-10 contains 144 million nucleotide bases and carries 983 genes. The Chromosome-11 contains 144 million nucleotide bases and carries 1,692 genes. The Chromosome-12 contains 143 million nucleotide bases and carries 1,268 genes. The Chromosome-13 contains 114 million nucleotide bases and carries 496 genes. The Chromosome-14 contains 109 million nucleotide bases and carries 1,173 genes. The Chromosome-15 contains 106 million nucleotide bases and carries 906 genes. The Chromosome-16 contains 98 million nucleotide bases and carries 1,032 genes. The Chromosome-17 contains 92 million nucleotide bases and carries 1,394 genes. The Chromosome-18 contains 85 million nucleotide bases and carries 400 genes. The Chromosome-19 contains 67 million nucleotide bases and carries 1,592 genes. The Chromosome-20 contains 72 million nucleotide bases and carries 710 genes. The Chromosome-21 contains 50 million nucleotide bases and carries 337 genes. Chromosome-22 contains 56 million nucleotides and carries 701 genes. Finally, the sex chromosome of all females called the (X) contains 164 million nucleotide bases and carries 1,141 genes. The male sperm chromosome contains 59 million nucleotide bases and carries 255 genes.

If you add up all genes in the 23 pairs of Chromosomes, they come up to 26,808 genes and yet we keep on mentioning 24,000 genes needed to keep us function normally. Out of 24,000 gene, 16,000 are good genes which keep us healthy and 6,000 are mutated or bad genes which are responsible for six thousand different diseases and 2,000 pseudogenes which has lost their functions. As I said above, a gene codes for a protein, not all 24,000 genes code for proteins. It is estimated that less than 19,000 genes code for protein. Because of the alternative splicing, each gene codes for more than one protein. All functional genes in our body make less than 50,000 protein which interact in millions of different ways to give a single cell. Millions of cells interact to give a tissue, hundreds of tissues interact to give an organ and several organs interact to make a human [1-6].

The purpose of describing the sequence of the entire human genome is to map, identify and locate all genes and move any gene from animal genome to plant genome including edible vegetables to create a complete vegetarian's diet. Sequencing Human Genome also help develop new tool kit of enzymes, the molecular scissors, to cut, paste and copy any piece of DNA from one species and insert to another species including animal genes to plants genome.

The completion of the Human Genome Project helps us follow the selective genetic breeding by sequencing egg and sperm and by discarding defected eggs and sperms. Some conservatives' members the followers of the Davenport's philosophy of our society will not accept the new discoveries. It is the parents who make the decision. After examining the sequence of their egg and sperm, the future parents should decide if they would use the defected egg and sperm, or should they discard the defected egg and sperm and use healthy egg and sperm which are free from any abnormal mutations. Despite these precautions, what if the sequence of the fertilized ovum shows the presence of an additional Chromosome-21 responsible for cause Mongolian disorder (Down Syndrome) who are unlikely to survive past thirty years. Should we allow the nature to take its course and let them die or should we bring them into this world by providing medical intervention and prolong their life, even though they will not live a quality life. Do you know that some handicapped children in

America are suing their parents for bringing them into this world where they become burden on society? Simple economy works here. The cost of medical treatment is unaffordable. May be some handicapped children will have to sue their parents in our country that will teach their parents a lesson.

Should we set up committees to advise parents and to draw guidelines for medical professionals so that they will make a rational judgment to advise parent who will determine if child A receives the precious treatment and will live, and child B will not receive the treatment and therefore will die. We need new ethical principles based on modern science. This is the main thrust of my arguments. The old ethical principles also came from people's head, but they were based on the information available to our elders long before the Human Genome Project was completed. Most ethical principles we used today were developed by Socrates about 2,500 years ago and everything that is written in philosophy since then is a footnote to his work. Although we have made a little progress in philosophy, we have made tremendous progress in genomic science. We are developing genomic medicine to keep people alive past one hundred years. Based on the genetic make-up, we are developing novel drugs to treat old age diseases such as Alzheimer, Cardiovascular diseases, and Cancers.

Genes are the unit of inheritance. As I said above, out of four-letter text, that is A-T and G-C, and three letters code for an amino acid called the Codon. The starting Codon in a gene is the Codon AUG which codes for amino acid Methionine. Long chain of DNA synthesis begins. The starting Codon is followed by a series of hundreds of Codons which codes for different amino acids in different species. There are three Stop Codons, and they are UAG, UGG, and UGA. Once the stop codons appear, DNA synthesis stops. Bacteria and Viruses have short codon chain. The longest chain is in a gene of Ducharme Muscular Dystrophy, a neurological disease whose chain extends to two and a half million codons. Once a gene is identified, using Restriction Enzymes, like EcoRI, we can cut, paste, and copy all genes individually making a Restriction Site map. Once a single gene is isolated and stored in cell libraries, we could compare the sequence of this gene with the Reference Sequence and the Thousand Genome Project to identify rare mutations responsible for genetic diseases. Sequencing genome is like extracting Gold from its Ore. Darwinian evolution by Natural Selection requires millions of years to accommodate the environmental conditions. Genetic Revolution liberates us from the shackles of the natural selection. In the Lab we can accomplish evolutionary changes in any species' genomes in days by cutting, pasting, and copying genes.

Once the diagnostic tests confirm the location of mutated genes for either monogenic or poly genic diseases such as cancers or cardiac diseases or Alzheimer; we could design drugs to shut off those genes. The greatest challenge is to shut off those mutated genes that are responsible for causing cancers.

### How to design drugs to shut off genes that Cause Cancers?

We design drugs to shut off genes that cause cancers. Professor WCJ Ross is one of the most distinguish English Scientists in the world. He is a Professor of Chemistry at the London University and Head of the Department at the Royal Cancer Hospital, a post-graduate medical center of the University of London. I am honored to be his graduate student, post-doctoral fellow and a special assistant and spent almost ten years in his Laboratory.

It was Professor Ross who worked on the development of anticancer drugs. An extremely lethal chemical called Nitrogen Mustard was tested against experimental animal tumor as an anti-cancer agent. We used the same method to make Nitrogen Mustard as was developed by German scientists during WWI. Nitrogen Mustard are the deadliest

class of chemicals. It was extensively used during WWI. It is believed that more than 20 million people died during WWI. In 1919, there was an epidemic of Flu. It is believed that the use of Nitrogen Mustard during WWI mutated a normal Flu virus to become a deadliest strain. People have no immunity against this strain. The flu killed more people around the world than the people died during the hostility of the WWI.

### Historical Background for Using Nitrogen Mustard for Treating Cancer

Fitz Heber, a German Army officer, worked on the development of Chemicals as a Weapon of War. He was responsible for making deadly Nerve gases and Nitrogen Mustards. Before the WWI, he was honored with a Nobel Prize for capturing Nitrogen for making Nitrate fertilizers directly from the atmosphere by burning the element Magnesium in the air forming its Nitride. Upon hydrolysis, Nitride is converted to its Nitrate. Using this method, we could make unlimited amount of fertilizer. Nitrate is also used for making explosive. Soon after the WWI, Heber was charged with a crime against humanity for releasing hundreds of cylinders of Chlorine gas on the Western front killing thousands of soldiers in the trenches. When Allied forces reached his residence, his son shot himself and his wife committed suicide. Heber went in hiding in Swiss Alps. After the War, German Government got his release as a part of the peace negotiations. Heber returned home to hero's welcome. Although he promised never to work on the chemical weapons again, secretly he continued to develop more lethal analogs of highly toxic chemicals like Nitrogen Mustards. It was Heber who first made the notorious Bis-dichloro-ethyl Methyl Amine. Because it smells like Mustard seeds, it is called as Nitrogen Mustard. During the next 20 years, before the beginning of the WWII, hundreds of more toxic analogs of Nitrogen Mustard were developed. The bad news is that they are highly toxic, and the good news is that they shut off genes.

Nitrogen Mustard was mercilessly used during the WWI by both German and Italian Armies against Allied forces. Most soldiers exposed to Nitrogen Mustard were freeze to death. Their blood analysis showed a sharp decline in Mitochondria and White Blood Cell (WBC). Since patients with the cancer of the blood called Leukemia, showed a sharp increase in WBC, Professor Ross and his group at the London University wondered if minimum amount of Nitrogen Mustard could be used to control Leukemia in cancer patients. It was a success. During the following 30 years, Ross developed hundreds of derivatives of Nitrogen Mustard to treat a variety of cancers. His most successful drugs are Chlorambucil, Melphalan and Merophan [7-11].

Over ten-year period, I made for Professor Ross dozens of analogs of Nitrogen Mustards. The deadliest among them was the Phenylenediamine Mustard. We use these compounds to check the sensitivity of the Tumors in the Tumor Bank. If tumors in the Tumor Bank become resistant, we must replace resistant tumor cells with fresh more sensitive tumors for testing other compounds.

### Rationale for Developing Nitrogen Mustard Analogs as Anti-Cancer Drugs

As I said above, I made several Nitrogen Mustards for Professor Ross. I will describe you how to make the Nitrogen Mustard by using Haber's crudest method. Haber reacted Methylamine with Ethylene oxide to make 2-bis dihydroxy ethyl methyl amine. It was chlorinated by heating with Phosphorus Penta Chloride in the Phosphoric Acid. If you noticed a faint smell of Mustard Seed, Congratulations, you got Nitrogen Mustard. If you are careless, you could be dead. You cool the solution and dilute with ice cold water; the oil floating in the aqueous solution was extracted with Chloroform. The solution is dried, and Hydrogen chloride gas is passed through to make its Hydrogen-Chloride salt. Nitrogen Mustard Hydrogen Chloride salt is separated. No matter how much precautions you take, after the experiment, if you would take an alcohol swab of walls, doors, knobs and run a mass spectra of alcohol extract, you find a spectral line corresponding to

Nitrogen Mustard. If you are exposed to Nitrogen Mustard and cross the threshold level, your WBC drops sharply and the energy providing Mitochondria die and you are most likely to freeze to death. Someone in the Defense department may make it, now a day, will anyone approve this study in the University Research Lab, probably No one. Your IRB (Institutional Review Board) and the safety committee will reject your proposal; and who will provide the funds for such an expensive study. The drug sensitivity between normal cell to cancer cell gives a ratio of toxicity called the Chemotherapeutic Index (CI). The larger the ratio the drug is more toxic to cancer cell. When tested against Walker Carcinoma 256 in Rats, most Nitrogen Mustards analogs cross-link both strands of DNA and give a CI of ten [12,13].

### Rationale for Developing Aziridine Analogs as Anti-Cancer Pro-drugs

Radio labeled study showed that Nitrogen Mustard shut off genes by binding to DNA by cross-linking both strands. It gives a Chemotherapeutic Index (CI) of ten which means that the drug is ten times more toxic to cancer cell when compared to normal cell. Higher TI means, it is more toxic to the cancer cells. We also discovered that radio-labeled Nitrogen Mustard does not bind to both strand of DNA simultaneously. First, one arm of the Nitrogen Mustard binds to a single strand of DNA, the Carbonium ion of the second arm is so reactive, it attacks its own Nitrogen atom forming a three-member intermediate ring called the Aziridine ion. Aziridine analogs serves as prodrug and are extremely unstable in the acidic medium. As the living cells grow, they use glucose as a source of energy. Glucose breaks down to Lactic Acid which opens the Aziridine ring generating a Carbonium ion which attacks a single strand of DNA causing mutation preventing cell division. The cancer cell dies. We made a series of Dinitro Phenyl Aziridine compounds to test against the experimental tumor Walker Carcinoma 256 in Rats. One compound the benzamide of Dinitrophenyl Aziridine (CB 1954) gave the CI of 70 highest ever recorded [14].

As I said above, in the Laboratory of Professor Ross, I had worked for almost ten years with the deadliest Nerve agents making their derivatives such as Nitrogen Mustards, Carbamates and Aziridines developed during Hitler's time for evil purposes. We converted the evil chemicals into good chemicals. These agents easily pass-through various layers of our skin from Ectoderm to Mesoderm to Endoderm. They easily enter the cell nucleus destroying the beta and gamma cell which develop immunity. Then they enter the nuclear membrane where they find the stem cells. Stem cells differ from say skin cells. In Stem cells all 24,000 genes are functioning, cells have not yet differentiated. On the other hand, differentiated cells like skin cells which are differentiated, the Epigenetic groups such as methyl group or Acetyl group have shut off all other genes except the skin cell genes.

While Professor Ross worked with the Nitrogen Mustard by cross-linking both strands of DNA, as his Doctoral student, I was assigned to work with Aziridines which binds to a single strand of DNA when activated in acidic medium. As a part of my Doctoral Thesis, I attached alkylating Aziridine to dyes like Dinitro Benzamide to attack the DNA of an experimental animal tumor called Walker Carcinoma 256. Aziridines analogs act as prodrugs. They remain inactive in neutral and basic media but activated in the Acidic medium. As I said above, the cancer cells grow faster than normal cells, they use more Glucose as a source of energy. Glucose breaks down to produce Lactic Acid. The Aziridine moiety is unstable in acidic solution. The Aziridine breaks down to open its ring to produce a positive Carbonium Ion. The Carbonium ion is extremely reactive; it binds to a single strand of DNA. It preferentially binds to N-7 of Guanine killing the tumor cells. Professor Ross and I have demonstrated the attack on N-7 of Guanine using the radio labeled studies. Over the years, I made 120 Dinitro-Benzamide derivatives for testing against Walker Carcinoma 256 in Rats [15,16]. When I introduced Aziridine and Carbamate moieties

to the Dinitro benzamide moiety, the compound was so toxic that its toxicity could not be measured. This line was discontinued at the London University, England, but the work was continued in America.

From our Labs at the Royal Cancer Hospital, University of London, England, I had sent to NIH (National Institutes of Health), America, over 120 drugs for NCI (National Cancer Institute) screening program. NCI honored me with the Fogarty International Fellowship Award to come to America to continue my work with Aziridines translating the animal work in Humans. As I said above, NIH is the largest biomedical center in the world. It has unlimited facilities (chemicals, equipment, and technical personnel). Twenty-one thousand best and brightest scientists selected from Ivy League schools from all over the world work in 26 institutes in more than three thousand labs. I was honored to join this group at NCI.

I developed the same rationale to continue my work in America. I brought the idea from London University of attacking one strand of DNA using Aziridine analog as prodrug, but I do not want to use the same dye Dinitro benzamide. One day, I came across a paper which described that methylated radio labeled Quinone crossed the Blood Brain Barrier. The X-ray showed that the entire radioactivity was concentrated in the Brain. I knew that Glioblastoma multiforme, the brain tumor, is a solid aggressive tumor like Walker Carcinoma in Rats. I decided to use Quinone moiety as a carrier for Aziridine rings to attack Glioblastoma. I was delighted when I realized by introducing just one Aziridine and one Carbamate moiety to Dinitro Benzene ring, by using Quinone, I could introduce two Aziridines and two Carbamates moieties to produce an extremely toxic diaziridine, dicarbamate Quinone, I named it AZQ, to attack Glioblastoma. The tumor not only stop growing, but also it started shrinking [17,18,19].

Over the years, I made several analogs of AZQ. My major concern was how toxic these compounds would be to the brain cells. Fortunately, brain cells do not divide, only cancer cells divide. AZQ and its analogs all act as prodrug. As I said above, to grow, cancer cells use glucose as a source of energy. Glucose is broken down to produce lactic acid. It is the acid which activates the aziridine and carbamate generating Carbonium ions attacking Glioblastoma. Over the years, I conducted over 500 experiments which resulted in 200 novel drugs which were tested against experimental animal tumors. Forty-five of them were considered valuable enough to be patented by US Government (US Patent 4,146,622 & 4,233,215). AZQ acts as a silver bullet. Glioblastoma was not only stop growing, but also start shrinking. For the discovery of AZQ, I was honored with the "2004 NIH Scientific Achievement Award" one of America's highest award in medicine and I was also honored with the "Vidya Ratna" a Gold Medal, one of India's National Medal of Honors (**Exhibit # 1,2,3 & 4**).

### Microbial Genome

Since the completion of the Human Genome Project just over eighteen years ago, we have made enormous progress. We have developed technologies to sequence the book of life faster and cheaper. We identified functional elements of the genome, the evolution of genome and the basis of cancers. We have not only sequence Eukaryote's genomes, but also Prokaryote's genomes. Today, we have sequenced the genomes of 250 Eukaryotes and more than 5000 genomes of Prokaryotes including bacteria and viruses. Several thousand genomes are being sequenced right now around the world. The Prokaryotes genomes are not enclosed in the nuclear membrane. The Prokaryotic cell has a single double stranded circular chromosome made of 4.6 million nucleotide base pairs. The chromosome is floating within the cytoplasm. Inserting Chloroplast genome in Prokaryotes would be easier. The good news is that the single Prokaryotes double every twenty minutes (some takes up to 60 minutes). Exposed to Sunlight, the photosynthetic apparatus, chloroplast, would rapidly absorb Carbon dioxide to convert it to Carbohydrate and releasing Oxygen.

As a result of the completion of the Human Genome Project, we have developed the toolkit for genetic engineering. The toolkit contains enzymes (such as restriction enzyme to cut the DNA, to join enzymes or to ligase the DNA and region of DNA in non-coding region which acts as enhancers, promoters, regulators, switches etc.) to cut, paste and copy small and large fragments of DNA. We have identified and isolated more than three thousand restriction enzymes like EcoRI which act as molecular scissors for cutting and pasting the Chloroplast genome in non-photosynthetic plants. These transgenic Chloroplast Genomes could be harvest on massive scale. We have sequenced the genomes of hundreds of living creatures. Sequencing Chloroplast genomes has provided a path to mitigate the climate change.

### Chloroplasts Genome

Chloroplasts are organelles present in plant cells and some eukaryotic organisms. Chloroplasts are the most important plastids which is a major double-membrane organelle found in the cells of plants and algae. Plastids are the site of manufacture and storage of important chemical compounds used by the cell. It is the structure in a green plant cell in which photosynthesis occurs. It is a primary site for splicing essential amino acids Codons.

Chloroplast is one of the three types of plastids. The chloroplasts take part in the process of photosynthesis, and it is of great biological importance. Animal cells do not have chloroplasts, but they have Mitochondria. All green plant take part in the process of photosynthesis which converts Carbon dioxide into carbohydrates its food in the presence of sunlight energy and the byproduct of the process is Oxygen that all animals breathe. This process happens in chloroplasts. The distribution of chloroplasts is homogeneous in the cytoplasm of the cells and in certain cells chloroplasts become concentrated around the nucleus or just beneath the plasma membrane. A typical plant cell might contain about 50 chloroplasts per cell.

The entire nucleotide sequence of Chloroplast Genomes has been determined. It is found to contain 120-190 thousand nucleotide base pairs. While a typical plant cell might contain about 50 chloroplasts per cell, most land plant chloroplast genomes typically contain around 110-120 unique genes. Some algae have retained a large chloroplast genome with more than 200 genes, while the plastid genomes from non-photosynthetic organisms may retain only a few dozen genes. Since some cells can survive without Chloroplast genes and others can survive with larger number of Chloroplasts genome without any harmful effect to cells, we wonder if we could introduce large number of Chloroplast genes splicing with essential amino acid codons in non-chloroplast living cells. We could insert the photo synthetic abilities in non-chloroplast cells to remove excess amount of Carbon dioxide from the atmosphere at the same time enriching them with important proteins. Trees perform the same photosynthetic function, but at a much slower rate. The microbial life form replicates much faster. The microbial genome doubles every 20 minutes. Large scale transgenic microbial or plants cells carrying genetically engineered Chloroplast hybrid could be harvested for public distribution. Since we all contribute to the Greenhouse gases, we all should participate in spreading synthetic Chloroplast Genome on ocean surface world-wide. Seventy-five percent of our planet is covered with water. Filamentous life forms or Algae floating on the water surface could be collected process to make supplemental food products. With global effort, may be within a hundred year, we might be able to reduce the level of Carbon dioxide to the pre-industrial or acceptable level.

### Genetic Engineering of Chloroplast Genome

One of the greatest intellectual achievements of the 21st century is the Genetic Engineering. We developed methods to cut these genes from the genome of one species, paste these genes into the genome of a second species and copy these genes into the genome of a third

species or to store this information in a Gene Library (collection of large number of identical genes) in living organisms. We also learn to scale up their product proteins into massive quantities so that we could supply these proteins to anyone around the world who needs it. A whole series of discoveries resulted in the development of the genetic engineering. We discovered the restriction enzymes which serve as molecular scissors to cut the DNA at a specific site then we discover enzyme DNA ligase to paste the genes then we also discover a series of enzymes such as proteases, DNA polymerase, RNA polymerase, reverse transcriptase which serves as catalyst to modify genes. Because of the development in genetic engineering, we were able to splice human insulin gene in yeast which is grown in huge bioreactor to produce insulin on a massive scale and were able supply to about 300 million diabetic around the world. Now, it is the time to do the same with the transgenic chloroplast genome for worldwide distribution. Genetic Engineering involves three steps:

#### Step One:

**Restriction Site:** It involves cutting out the genes from the chromosome at specific site of the genome using the restriction enzymes which serve as molecular scissors. More than two hundred restriction enzymes have been discovered for cutting DNA at different sites and at different lengths. DNA fragments are purified by electrophoresis and sequenced their base pairs. The gene present within these fragments cannot be used to make proteins because the enzymes present in the host cell will break down to pieces. These fragments must be protected by recombining with the DNA of other species.

#### Step Two:

**Recombinant Technology:** The DNA fragments produced by restriction enzymes are protected by recombining with the DNA of the following three species which serve as Vectors (a) Viruses, (b) Plasmids, or (c) Chloroplast. To accommodate larger fragments of DNA, we have made Phagemid, Cosmids, BAC (Bacterial Artificial Chromosome and YAC (Yeast Artificial Chromosome). Same restriction enzymes are used to cut fragments from vectors as well. Enzyme ligase joins these fragments. Easy purification methods are developed. For example, we used the plasmids which carry antibiotic resistant genes such as ampicillin or tetracycline. After ligation, ampicillin or tetracycline is added to eliminate all plasmids which do not carry ampicillin or tetracycline resistant genes.

#### Step Three:

**The Host Cell Amplification:** One of the greatest challenges is to scale up the desired product of the genes into large quantities. How could we transfer the recombined vectors into a host cell to clone or make large number identical copies of the same proteins? Stanley Cohen was able to transfer victor plasmids into the host cell for amplification by simply adding Calcium salts such as Calcium Phosphate to the solution which enlarges host cell pores and allows hundreds of recombined Plasmids to replicate in host cells. The purified gene can be saved in living organism as Gene Library (collection of large number of identical genes). Libraries for all 24,000 genes can be stored for future use. Genetic Engineering will open new frontiers for developing new drugs for treating old diseases.

We are training thousands of scientists around the world to master the techniques of Genetic Engineering. There are three million known and thirty million living species on Earth. Using restriction enzymes, they are learning to cut DNA of various lengths of various species and preparing the restriction site map of various species. Using recombinant technology, they are protecting these fragments of DNA by preparing Vectors in viruses, plasmids, or chloroplasts genomes. Making Clones (identical copies) of the same DNA fragments in an autonomously replicating host cells to sequence the colons and to make libraries (collection of large number of the same colons) of all 24,000 genes, good and bad both from human genome. Good genes are used

to produce useful proteins to treat various diseases such as insulin to treat diabetics. On the other hand, bad genes (such as oncogenes) are used to induce tumors in animals to test novel anti-cancer drugs.

As I said above, Chloroplast Genome has been sequenced. The good news is that both Prokaryote and some Eukaryotes carry Chloroplast. Using recombinant technology, we could make transgenic hybrid carrying Chloroplast gene in the TI Plasmids (Crown gall) is caused by *Agrobacterium tumefaciens*, a Gram-negative, bacilliform bacterium that is normally associated with the roots of many different plants in the field. The Tumor Induced Plasmids (TI Plasmids) found in the gall (plant tumors have the unique ability to replicate rapidly like cancer cells), they could be used to splice chloroplast genome to produce transgenic Prokaryotes for world-wide distribution.

As I said above, the Chloroplast Genome will carry essential amino acid genes which will produce a new kind of nutritious food for the burgeoning population of the world. We will splice essential amino acid genes in the Chloroplast Genome, it will not only remove CO<sub>2</sub> from the atmosphere, but also it will produce Oxygen and essential amino acid rich carbohydrate compounds which could be purified and used for human consumption. What are the Essential Amino Acids and why we need them? Proteins in our body is made of twenty amino acid. Our body makes all amino acids except the following eight which are called essential amino acids and they are: Valine, Leucine, Isoleucine, Phenylalanine, Tryptophan, Lysine, Histidine, and Threonine. We need outside source like meat. Without the essential amino acids, we develop a variety of diseases. Can we get essential amino acids in our diet without eating meat? Essential amino acids Codons are identified. We can insert these codons in the Chloroplast Genome of the microbial plant life which could serve as food and food products. The Codons for each essential amino acid and their alternative codons are described below: Valine (GTT, GTC, GTA, GTG), Leucine (CTT, CTC, CTA, CTG; TTA, TTG), Isoleucine (ATT, ATC, ATA), Phenylalanine (TTT, TTC), Tryptophan (TGG), Lysine (AAA, AAG), arginine (CGT, CGC, CGA, CGG; AGA, AGG), Histidine (CAT, CAC), Methionine (ATG), Threonine (ACT, ACC, ACA, ACG).

### Gene Editing using CRISPER/ Cas9 Technology

Among the most recent developmental technique in genetic engineering is the gene editing called CRISPER referred to a region of bacterial DNA whose acronym stood for "Clustered Regularly Interspaced Short Palindromic Repeats." Unfortunately, gene editing could not be used to splice Chloroplast Genome in bacterial cells. We still need to use recombinant technology to cut paste and copy Chloroplast Genome in bacteria. Unlike GMO (Genetically Modified Organisms) species where a new gene is inserted by a random process to introduce a novel protein which gives organism a beneficial trait, no new gene is inserted in gene editing, but a tiny change is made to correct a spelling error of nucleotides in the existing genes. DNA editing is done when a specific sequence of RNA fragment is attached to a specific protein called Cas9 (This protein also acts as a general molecular scissor, but different from restriction enzymes which acts as a specific scissor for each restriction site) forming a RNA-protein complex called CRISPER/Cas9. To edit a gene carrying a mutated nucleotide, the RNA complex is hybridized with the muted DNA, which matches the complimentary sequence. Once it is hybridized, the CRISPER cuts the double stranded DNA at both sides and replaces it with the correct RNA and using an enzyme ligase to join at both sides. CRISPER technology helps us edit out or remove a single spelling error or few letters or words sentences and paragraphs of genetic script. Once edited, the information is passed on to the next generation.

Restriction enzymes are proteins, and they act as scissors to cut the double stranded DNA at a specific site. Several hundred restrictions enzymes have been isolated to cut DNA which could be used to cut either small fragments or large fragments either single stranded or double stranded DNA. By trial and error, we must find the specific

restriction enzyme which could cut the Chloroplast Genome for making its recombinant variety for sequencing and making millions of clones in bacteria.

### Limitation of CRISPER

Studies are being conducted to use gene editing to treat CNS (Central Nervous System) diseases. CNS diseases are multi genetic in nature and they include Schizophrenia, Bipolar Disorder, Epilepsy, Parkinson Disease, Huntington Chorea etc. Using Gene editing to treat CNS diseases require precision and accuracy. A single nucleotide error could cause an irreparable damage in humans because editing error could not be corrected. Using chemicals to treat CNS diseases is much safer. If a drug being used for treating CNS disease found to be too toxic or harmful, it could easily be replaced with a less toxic analog. I have made 45 AZQ (US Patent 4,146,622 & 4,233,215) derivatives for treating CNS diseases like Glioblastomas. If one AZQ analog is found to be too toxic, it could easily be replaced with another analog.

As I said above, instead of using gene editing, we must use GMO type technology to splice Chloroplast Genome in Prokaryotes. Using restriction enzymes like EcoR1, we could cut the DNA at various sites of the chloroplast genome and recombine within the TI plasmids (Tumor induced plasmid obtained from the plant tumor). Once the hybrid is obtained, it could be cloned in millions. The hybrid Chloroplast clone could be transferred to both Prokaryotes and some Eukaryotes. The transgenic Chloroplast could be put to work on global scale to absorb and remove excessive amount of atmospheric Carbon dioxide.

All seven continents could be used to harvest the Chloroplast transgenic microbial life forms. If life exists in extreme and harsh climate from Sahara to Antarctica, extremophile could be used to make transgenic Chloroplast which should function in extreme climate to absorb Carbon dioxide. For example, Antarctica is the 5th largest continent with 5 million square mile long surface and has a 3 miles thick sheet of ice. Besides, Penguins, Elks, Seals and Great Whales, most residents are Plankton, Krill and Algae. They could be put to work to create the transgenic Rainforest of Amazon for chloroplast genome.

What should the future generation of geneticists explore next? The answer is that they should work on the Chloroplast genome which all living green plant use to absorb Carbon dioxide to generate their food carbohydrate and Oxygen. They could splice Chloroplast genome in all rapidly growing plants and microbial life forms. Chloroplast genome is present in most Prokaryotes and some Eukaryote. Photosynthesis will not only reduce the pollutant Carbon dioxide but also produces Oxygen. We must continue to reforest our planet on massive scale. The Chloroplast Genome has already been sequenced. What has not been done is to move around the chloroplast genomes from cell to cell. Using restriction enzymes, we can cut paste and copy Chloroplast genomes from one living cell to another living cell. We can move Chloroplast genome from gene rich organisms to gene poor living cells. The next generation of geneticists should harvest Chloroplast rich genome plants and animals at global scale to distribute worldwide particularly in uninhibited areas of planet Earth like Antarctica. During the next hundred years, we could create Amazon Forest of Prokaryote chloroplast rich plants.

One of the great advantages of using Chloroplast technology is that it prevents genetic drift by preventing spores from spreading by winds and insects contaminating neighboring farms and fields by locking the trans-gene within the Chloroplasts genome.

To succeed in spreading transgenic chloroplast genome, we need to make a global effort. Since we all are responsible for contributing to Carbon dioxide, we all should participate in this global experiment to diatribe Chloroplast genome to reduce the level of Carbon dioxide.



Would this hundred-year experiment alter the atmospheric composition to bring down the level of Carbon dioxide to pre-industrial level? May be? Would the additional amount of Oxygen set massive forest fire as it did when the level of Oxygen was more than 20 percent in the early Earth? May be? But we can control the level of Chloroplast genome at will. We are conducting a 100-year experiment to reduce the level of Carbon dioxide and increase Oxygen. We could not only control the level of Carbon dioxide, but also stop and reverse the current trend anytime.

Lastly, what if we do nothing? Our children and our children's children will have to pay the price. We must ask ourselves; is there an impending environmental crisis? We look for evidence and require critical thinking to find the solution. It is up to each of us to look for evidence. The ravages of climate change have been observed worldwide and are on display in recent years in creating the super storms; massive tornadoes; there is a rise in sea level; and a reduction of icesheet and disappearance of glaciers; severity of tornados and super storms; the increase in the forest fire, intense droughts and deadly heat waves that will only get worse as the Carbon dioxide level increases. The same condition we will face tomorrow as the residents of Maldives are facing today. Because of the sea rise, the islands of Maldives are already sinking. Where would we move over a quarter million Greenhouse refugees, to Solomon Islands, or to Turkey or Indonesia? Would these countries welcome so many Green House refugees? Hardly likely.

### Conclusion

Now, it is possible to mitigate the impact of climate change by using the toolkit of biotechnology developed during the race to sequence and complete the Human Genome Project. We can teach the next generation of scientists how to cut, paste and copy Chloroplast genome in microbial life form for worldwide release. It is estimated that every person on Earth contributes about ten tons of Carbon dioxide per year to our atmosphere. Now, we can all do something to clean our environment. Every person on Earth must participate in distributing Chloroplast rich microbial life on all seven continents. May be within a hundred year, we might be able to bring back the level of Carbon dioxide to a more acceptable level. For the first time we learn that our destiny lies in our own hands.

### References

1. Watson JD, Crick FHC (1953) A structure for deoxyribose nucleic acid. *Nature* 171: 737-738.
2. Genome-Wide Identification of Long Non-Coding RNAs and Their Regulatory Networks *Nature* (2001) 409: 934-941.
3. International Human Genome Sequencing Consortium. Initial sequencing and analysis of the human genome (2001). International Human Genome Sequencing Consortium. Initial sequencing and analysis of the human genome. *Nature* 409: 860-921.
4. International Human Genome Sequencing Consortium (2004). *Nature* 431: 931-945.
5. Genomic sequence of the pathogenic and allergenic filamentous fungus *Aspergillus fumigatus* (2005). *Nature* 438: 1151-1156.
6. DNA Sequencing at 40 (2017). *Nature* 550: 345-353.
7. Chlorambucil - CancerConnect News". *CancerConnect News* (2015) 12-21.
8. Ross WCJ (1953) "The Chemistry of Cytotoxic Alkylating Agents" In *Advances in Cancer Research* by Greenstein JP, and Haddow A, Academic Press, Inc., New York 397-449.
9. Ross WCJ (1962) "Biological Alkylating Agents" Butterworth, London.

10. Ross WCJ (1949) *Journal of Chemical Society* 183.
11. Ross WCJ (1950) *J Chem Soc* 2257.
12. Ross WCJ, Mitchley BCV (1964) *Ann Rep Brit Empire Cancer Campn* 42: 70.
13. Melphalan *Lancet* 370: 1209-1218.
14. Cobb LM, Connors TA, Elson LA, Khan AH, Mitchley BCV, et al. (1969) "2,4-Dinitro-5-Ethyleneiminobenzamide (CB 1954): A Potent and Selective Inhibitor of the Growth of the Walker Carcinoma 256". *Biochemical Pharmacology* 18: 1519-1527.
15. AH Khan and WCJ Ross (1969/70) "Tumour-Growth Inhibitory Nitrophenylaziridines and related compounds: Structure-Activity Relationships" *PART I Chem-Biol Interactions* 1: 27-47.
16. Khan AH and Ross WCJ (1971/72) "Tumour-Growth Inhibitory Nitrophenylaziridines and related compounds: Structure-Activity Relationships" *PART II Chem-biol interactions* 4: 11-22.
17. A Hameed Khan and John Driscoll (1976) "Potential Central Nervous System Antitumor Agents: Aziridinybenzoquinones. *PART I Journal of Medicinal Chemistry* 19: 313-317.
18. Ed Chou, A Hameed Khan and John Driscoll (1976) "Potential Central Nervous System Antitumor Agents: Aziridinybenzoquinones. *PART II Journal of Medicinal Chemistry* 19: 1302.
19. "Aziridiny Quinone: Anti-transplanted Tumor Agents". UNITES STATES PATENT # 4,146,622. (March 27, 1979) Investors: John S. Driscoll; A. Hameed Khan; Feng-e-Chou, NIH, Maryland, USA.

### Exhibit # 1

2004 NIH Scientific Achievement Award Presented to Dr. Hameed Khan By Dr. Elias Zerhouni, The Director of NIH During the NIH/APAO Award Ceremony held on December 3, 2004.



Dr. Khan is the Discoverer of AZQ (US Patent 4,146,622), a Novel Experimental Drug Specifically Designed to shut off a Gene that causes Brain Cancer for which he receives a 17-year Royalty for his invention (License Number L-019-01/0). To this date, more than 300 research papers have been published on AZQ. The award ceremony was broadcast live worldwide by the Voice of America (VOA). Dr. Khan is the first Indian to receive one of America's highest awards in Medicine.

### Exhibit # 2

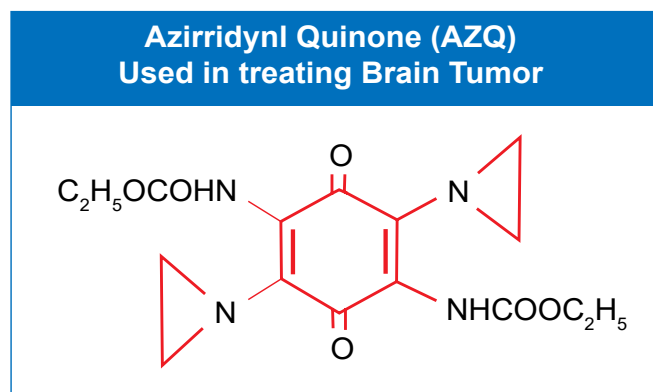
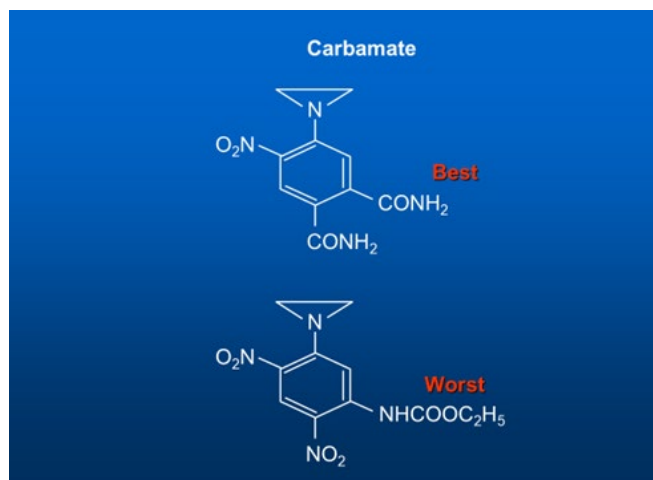
His Excellency, Dr. A.P.J. Abdul Kalam, The President of India Greeting Dr. A. Hameed Khan



Discoverer of anti-cancer AZQ, after receiving 2004, Vaidya Ratna, The Gold Medal, One of India's Highest Awards in Medicine At The Rashtrapathi Bhavan (Presidential Palace), in Delhi, India, During a Reception held on April 2, 2004.

### Exhibit # 3

Single Strand DNA Binding Aziridine and Carbamate



U.S. Patent 4,146,622

### Exhibit # 4

Gold Medal for Dr. Khan



Dr. A. Hameed Khan, a Scientist at the National Institutes of Health (NIH) USA, an American Scientist of Indian Origin was awarded on April 2, 2004. Vaidya Ratna, The gold Medal, one of India's Highest Awards in Medicine for his Discovery of AZQ (US Patent 4,146,622) which is now undergoing Clinical Trials for Treating Brain Cancer.