SOME BACKGROUND FOR GENETIC ENGINEERING

by

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In just the last few years we have seen the appearance on our planet of new living creatures, rebuilt by man from their conventional models, designed to perform tasks which will benefit man. This new biologic technology has the promise of improving vastly our medical capability, our food supply, even the quality of human individuals.

It all stems from our recent knowlege about the genetic system and its environment in the living cell. Now we know how to change the genetic structure which governs the operation of the cell, making the cell produce materials different from any they had produced before. And since the production of these highly complex organic compounds is far more efficient in the environment of the cell than it is in the environment of a laboratory, or a ~~chemical plant~~, these new materials carry a far cheaper price tag ~~than if we had produced them in typical chemical plants, starting the process with basic raw materials~~.

Human insulin is now beginning to be produced in this manner. A more exciting material, the human antibody, leucocyte interferon, which has stopped the growth of certain tumors and then drastically shrunk the size of these tumors--this antibody has been produced by inserting a human gene into the genetic code of a bacterium. Previously, this antibody, as refined laboriously from whole blood had carried a price tag of $200,000 for a 100 ml. bottle, the size bottle that will fit inside your hand.

Plant breeders say they feel confident of altering the gene structure of the corn plant to accommodate the genes from a legume, such as a bean or a pea, which would allow this corn to form nitrogen nodules on its roots and thus not require the huge amount of nitrogen fertilizer now needed to give a satisfactory yield of grain.

Animal breeders are working on their genetic machinery to develop disease-resistant strains and breeds with improved muscle structure, thus improved cuts of meat.

Generally, everything sought after in the way of biological improvement should be speeded up by many orders of magnitude. Instead of patiently crossing one strain to another through countless time-requiring generations of offspring to obtain a particular characteristic, it should be possible soon to locate that portion of gene structure which governs, and concentrate on altering this amount of chemistry only, in order to produce the new desired individual. Already the total genetic code of many simple cells is stored in computers.

To say that something is possible with available technology is indeed not to say that it is simple or quickly attainable. Let us consider some of the elements involved.

This series of drawings was produced by the little-known scientific illustrator, Madamoiselle Marie Wooldridge, working from photomicrographs. This first one represents magnification on the order of 200,000 times, or about as far as we can go with the electron microscope. On this scale an ant would be a half of a mile long. The drawing demonstrates the

great similarity between plant and animal cells as far as the components are concerned, the differences being mainly in shape and density. In humans, there are about 3 billion new cells produced each minute, to replace the same number which die. We do not know why cells die. White blood cells live about 13 days; red cells about 120 days. Liver cells live Some about 18 months. Nerve cells can live over 100 years, but they lose the ability to reproduce themselves after they mature.

In the cell nucleus are the chromosones, 46 in every human cell, whether muscle cell, blood cell, liver cell, or which (except the sex cells). In the chromosomes are the DNA units, again the same number in each type of cell. You may recall a stimulating paper which I delivered to this society several years ago describing the cloning of a frog, in which the egg was fertilized by the chromosomes in a skin cell of the same individual.

How does the cell do its business of turning out highly complex protein structures. We'll use the next two drawings. In the cell nucleus, the DNA unit is untwisted and seperated by an ensyme, allowing the messenger RNA to line up along one strand of the DNA helix. (Point this out) The messenger RNA thus picks up the sequence of amino acids needed to form the particular protein that the DNA wants formed. The messenger RNA thereafter leaves the nucleus and goes to an area in the cell called a ribosome, stretches itself out along the surface of this material, and waits for the transfer RNA to bring the amino acids which will fit in the coded equence along its length. Transfer RNA units are comparatively simple, three-element coded structures which stay around the ribosomes. Their only function is to transport a particular amino acid to its exact position in the lineup on the messenger RNA. There are twenty types of

this transfer RNA, corresponding to the twenty common amino acids found in proteins. When the sometimes hundred or more amino acids have been lined up according to the equence specified in the messenger RNA, the protein is complete. If it is not to be used internally, it is then secreted through the cell membrane. There is a code on the DNA strand for starting a particular protein and a code at the end to stop the sequence. These codes direct the formation of enzymes, the first of which chemically stimulates the process, the second of which forbids the process.

One especially baffling question is why cells differentiate. How does a developing embryo know when to start producing nerve cells, skin cells, and so on, considering that each cell supposedly has absolutely the same kind of, and the same amount of DNA material?

Now for the DNA unit itself. (Pass out photocopies) This is an atomic model of a structure which has never been observed like this in nature. The only known parameters were the X-ray measurements of width and density, plus the constituent chemical bases, when Francis Crick, of Cambridge, and James Watson, of Harvard, incredibly deduced this double helical arrangement which would accommodate chemically all the known data. This was in 1953. It has withstood experimental testing ever since.

The drawing you see is of only a segment of a living DNA strand. The real thing might show repeats of this segment, plus segments in which the placement of some of the groups were interchanged. On the scale used in this drawing, a DNA strand in a complex species such as man

would be over a mile long. Human DNA is believed to have about 150,000 genes; that is, 150,000 discrete chemical messages, and as many as 300 million individual code letters. When the cell divides in reproducing itself, this double helix unwinds into the shape of a ladder, then seperates down the rungs of the ladder, each half then going toward its respective half of the dividing nucleus. The DNA in man is of course not the same as DNA in cattle, or in corn, or in a cactus plant because different chemical instructions are required for each.

In 1975, a class of enzymes was discovered which could fragment the DNA structure with precision, in some cases resulting in pieces as small as a single gene. The enzymes could break the twisted ladder of DNA at a different point on the two sides, making for what researchers called "sticky ends." The sticky ends of one DNA species were found to attach firmly to the sticky ends of another species, so that for the first time it was possible to produce a new species by changing the DNA code. DNA fragments from a toad frog and from a fruit fly were made to reproduce in a new species of the E. Coli bacterium. It was this work which gave rise to the terms "gene splicing" and "genetic engineering." It was this work also which won the first patent in the field, awarded jointly to Stanford University and the University of California. The patent was upheld in a close decision by the U.S. Supreme Court which overruled the ~~thesis~~ contention that living creatures were beyond the realm of patent rights.

One of the great advantages of using bacteria to produce wanted materials is the rate at which bacteria multiply themselves. The E. Coli, for example, divides one every 20 minutes. In 24 hours, this exponential progression has produced over 2 billion units.

With all this immense new knowlege of the workings of living cells, we may ask the old question, "How did this system begin? Is it possible that such complexity is the result of an evolutionary process, where incremental adjustments to a changed environment make such changes that a new species finally appears? Let us examine this.

One of the authorities in the field is a Russian named A. I. Oparin. He thinks that conditions upon the earth's surface have changed so much from the time when the molten mass of the earth first solidified and cooled that the early chemical reactions which may have started life are not going on any longer. When the earth had just cooled down, its surface was covered by carbides, compounds of metal and carbon. Carbides are very unstable substances: few are found naturally any more. Mr. Oparin's thesis is that these carbides reacted violently with water to form gases--methane, acetylene, and some more-complex hydrocarbons. The atmosphere, which was very different from our own, also contained ammonia, nitrogen, and oxygen, thus making for a total environment in which complex molecules, like proteins, might have formed. Of course, the mere presence of hydrocarbons and ammonia do not make the complex protein molecule. A powerful and exacting energy manipulation must also

occur. Mr. Oparin believes that sunlight, atomic radiation, and lightning discharges over a long period of time accomplished the miraculous combination. Later, and also by chance, more complex structures like DNA were formed. This DNA and a simpler group of proteins were brought together in just the right arrangement, still by chance, so that the DNA used the simpler proteins to form a protective sheath around itself and somehow learned to divide in the reproduction of itself.

Has anyone tried to test this theory experimentally? In the early 1960's a California group set up a closed environment of hydrogen, methane, ammonia, and water. The mixture was circulated for a week over a strong electric spark. When the analysis was made at the end of the week, three amino acids were found, three of the 20 building blocks of proteins. Since then, other research groups have used different energy sources--ultraviolet light and nuclear particles from an accelerator. They produced two more of the amino acids. But none of the experiments has produced all 20 of the amino acids, and none has produced a compound more complex than these, nothing close to synthesis of a protein.

Recall the early attempts to synthesize DNA. It was found that with all the protein factions present, it was not possible to start the DNA synthesis without a "primer" of complete, intact DNA. In 1972, an MIT group built a portion of a complete DNA structure, a portion that would form a single messenger RNA unit. They did this by building piece by piece, using the most sophisticated techniques, over a period of nine years. This seems hardly the sort of thing that might have occurred in nature, entirely by chance, even in nine trillion years.

What is fairly commonplace today is the taking of parts of existing DNA from one cell and inserting them into the broken portion of DNA in another cell. It does appear that an entire DNA structure could be synthesized if a massive effort were made, but it would prove little beyond the fact that the whole is indeed equal to the MIT parts. That it could be done using scores of technicians in a large laboratory over a substantial number of years only reinforces my feeling that it is so utterly improbable that such a delicate, intricate fabrication could have occurred in a natural environment entirely by chance--no matter how much atomic radiation, how much sunlight, or how many bolts of lightning had their effect. The DNA system, including its RNA subsystems, is just too complex.

I have a friend who has spent his life as a research chemist. He says "It's like having all the parts of an automobile laying out there on the ground, and waiting for strong winds or a series of twisters to somehow put them together."

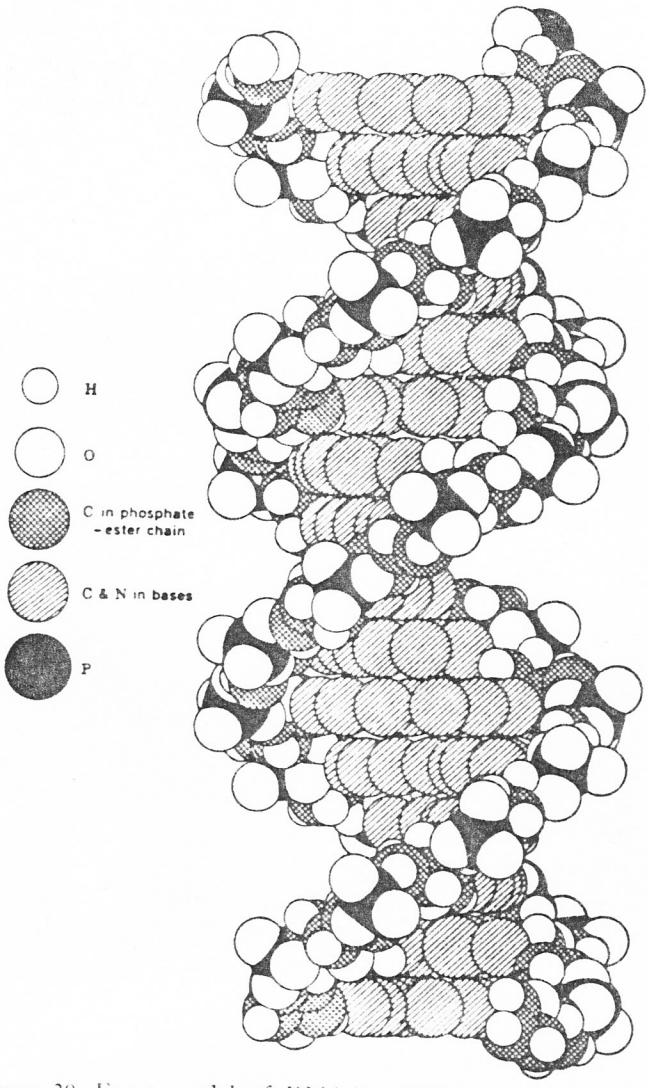

Figure 20. Exact model of DNA by Wilkins to refined data from X-ray crystallography. In this model each atom's sphere of influence is shown solid.

On this scale, complete human DNA would be over a mile long