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Introduction

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Introduction

Humankind is gaining an increasing understanding of heredity and variation among living things—the science of genetics. This report examines both the critical issues arising from the science and technologies that spring from genetics, and the potential impacts of these advances on society. They are the most rapidly progressing areas of human knowledge in the world today.

Genetic technologies exist only within the larger context of a maturing science. The key to planning for their potential is understanding not simply a particular technology, or breeding program, or new opportunity for investment, but how the field of genetics works and how it interacts with society as a whole.

The technologies that this report assesses can be expected to have pervasive effects on life in the future. They touch on the most fundamental and intimate needs of mankind: health care, supplies of food and energy, and reproduction. At the same time, they trigger concerns in areas equally as important: the dwindling supplies of natural resources, the risks involved in basic and applied scientific research and development, and the nature of innovation itself.

As always, some decisions concerning the use of the new technologies will be made by the marketplace, while others will be made by various institutions, both public and private. In the coming years, the public and its representatives in Congress and other governmental bodies will be called on to make difficult decisions because of society's knowledge about genetics and its capabilities.

This report does not make recommendations nor does it attempt to resolve conflicts. Rather, it clarifies the bases for making judgments by defining the likely impacts of a group of technologies and tracing their economic, societal, legal, and ethical implications. The new genetics will be influential for a long time to come. Although it will continue to change, it is not too early to begin to monitor its course.

The origins of genetics

For the past 10,000 years, a period encompassing less than one-half of 1 percent of man's time on Earth, the human race has developed under the impetus of applied genetics. As techniques for planning, cultivating, and storing crops replaced subsistence hunting and foraging, the character of humanity changed as well. From the domestication of animals to the development of permanent settlements, from the rise of modern science to the dawn of biotechnology, the genetic changes that mankind has directed have, in turn, affected the nature of his society.

Applied genetics depends on a fundamental principle–that organisms both resemble and differ from their parents. It must have required great faith on the part of Neolithic man to bury perfectly good grain during one season in the hope of growing a new crop several months later–faith not only that the seed would indeed return, but that it would do so in the form of the same grain-producing crop from which it had sprung. This permanence of form from one generation to the next has been scientifically understood only within the past century, but the understanding has transformed vague beliefs in the inheritance of traits into the science of genetics, and rule-of-thumb animal and plant breeding into the modern manipulations of genetic engineering.

The major conceptual boost for the science of genetics required a shift in perspective, from the simple observation that characteristics

passed from parents to offspring, to a study of the underlying agent by which this transmission is accomplished. That shift began in the garden of Gregor Mendel, an obscure monk in mid-19th century Austria. By analyzing generations of controlled crosses between sweet pea plants, Mendel was able to identify the rudimentary characteristics of what was later termed the gene.

Mendel reasoned that genes were the vehicle and repository of the hereditary mechanism, and that each inherited trait or function of an organism had a specific gene directing its development and appearance. An organism's observable characteristics, functions, and measurable properties taken together had to be based somehow on the total assemblage of its genes.

Mendel's analysis showed that the genes of his pea plants remained constant from one generation to the next, but more importantly, he found that genes and observable traits were not simply matched one-for-one. There were, in fact, two genes involved in each trait, with a single gene contributed by each parent. When the genes controlling a particular trait are identical, the organism is homozygous for that trait; if they are not, it is heterozygous.
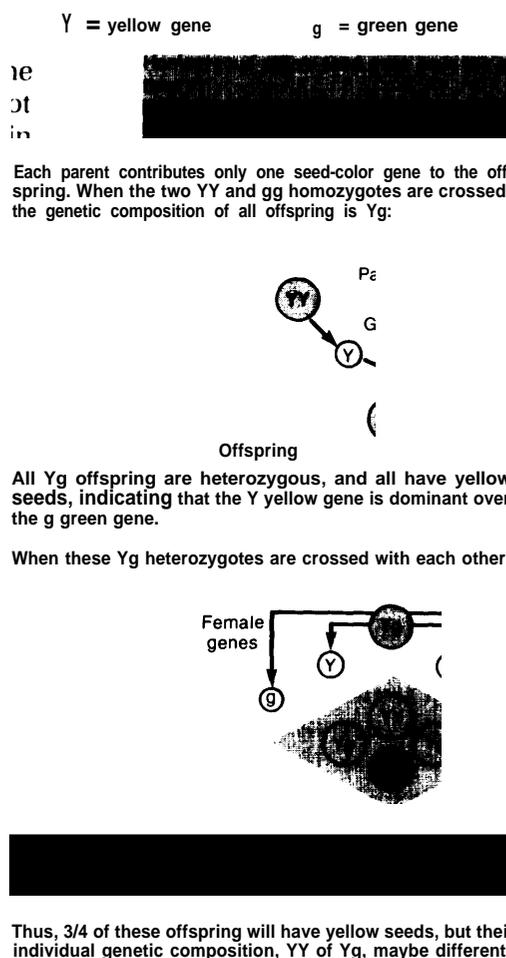
In the Mendelian crosses, homozygous plants always retained the expected characteristics. But heterozygous plants did not simply display a mixture of their different genes; one of the two tended to predominate. Thus, when homozy - gous yellow-seed peas were crossed with homozygous green-seed plants, all the offspring were now heterozygous for seed color, possessing a "green" gene from one parent and a "yellow" from the other. Yet all of them turned out to be indistinguishable from the yellow-seed parent: Yellow-seed color in peas was dominant to green.

But even though the offspring resembled their dominant parent, they could be shown to contain a genetic difference. For when the heterozygotes were now crossed with each other, a certain number of recessive green-seed plant again appeared among the offspring. This occurred whenever an offspring was endowed with a pair of genes that was homozygous for

the green-seed trait—and it occurred at a rate consistent with the random selection of one of two genes from each parent for passage to the new generation. (See figure 5.)

Genes were real—Mendel's work made that clear. But where were they located, and what were they? The answer, lay within the nucleus of the cell. Unfortunately, most of the contents of the nucleus were unobtainable by biologists in Mendel's time, so his published findings were ignored. Only during the last decades of the 19th century did improved microscopes and new dyes permit cells to be observed with an acuity never before possible. And only by the

Figure 5.-The Inheritance Pattern of Pea Color

Y = yellow gene   g = green gene

Each parent contributes only one seed-color gene to the offspring. When the two YY and gg homozygotes are crossed, the genetic composition of all offspring is Yg:

Offspring

All Yg offspring are heterozygous, and all have yellow seeds, indicating that the Y yellow gene is dominant over the g green gene.

When these Yg heterozygotes are crossed with each other:

Female genes

Thus, 3/4 of these offspring will have yellow seeds, but their individual genetic composition, YY of Yg, maybe different.

SOURCE: Office of Technology Assessment.

beginning of the 20th century did scientists *rediscover* **Mendel's work and begin to appreciate fully the significance of the cell nucleus and its contents.**

Even in the earliest microscopic studies, however, certain cellular components stood out; they were deeply stained by added dye. As a result, they were dubbed '(colored bodies, " or chromosomes. Chromosomes were seen relatively rarely in cells, with most cells showing just a central dark nucleus surrounded by an extensive light grainy cytoplasm. But periodically the nucleus seemed to disappear, leaving in its place long thready material that consolidated to form the chromosomal bodies. (See figure 6a.) Once formed, the chromosomes assembled along the middle of the cell, copied themselves, and then moved apart while the cell pinched itself in half, trapping one set of chromosomes in each of the two halves. Then the chromosomes themselves seemed to dissolve as two new nuclei appeared, one in each of the two newly formed cells. (See figure 6b.)

Thus, the same number of chromosomes appeared in precisely the same form in every cell of an organism except the germ, or sex, cells. Furthermore, the chromosomes not only remained constant in form and number from one generation to the next, but were inherited in pairs. They were, in short, manifesting all the traits that Mendel had prescribed for genes almost three decades earlier. By the beginning of the 20th century, it was clear that chromosomes were of central importance to the life history of the cell, acting in some unspecified manner as the vehicle for the Mendelian gene.

If this conclusion was strongly implied by the events of cell division, it became obvious when reproduction in whole organisms was analyzed. It had been established by the latter part of the 19th century that the germ cells of plants and animals—pollen and ovum, sperm and egg—actually fuse in the process of fertilization. Germ cells differ from other body cells in one important respect—they contain only half the usual number of chromosomes. This chromosome halving within the cell was apparently done very precisely, for every sperm and egg contained exactly one representative from each chromosome pair. When the two germ cells then fused during fertilization, the offspring were supplied with a fully reconstituted chromosome complement, half from each parent. Clearly, chromosomes were the material link from one generation to the next. Somewhere locked within them was the substance of both heredity—the fidelity of traits between generations; and diversity—the potential for genetic variation and change.
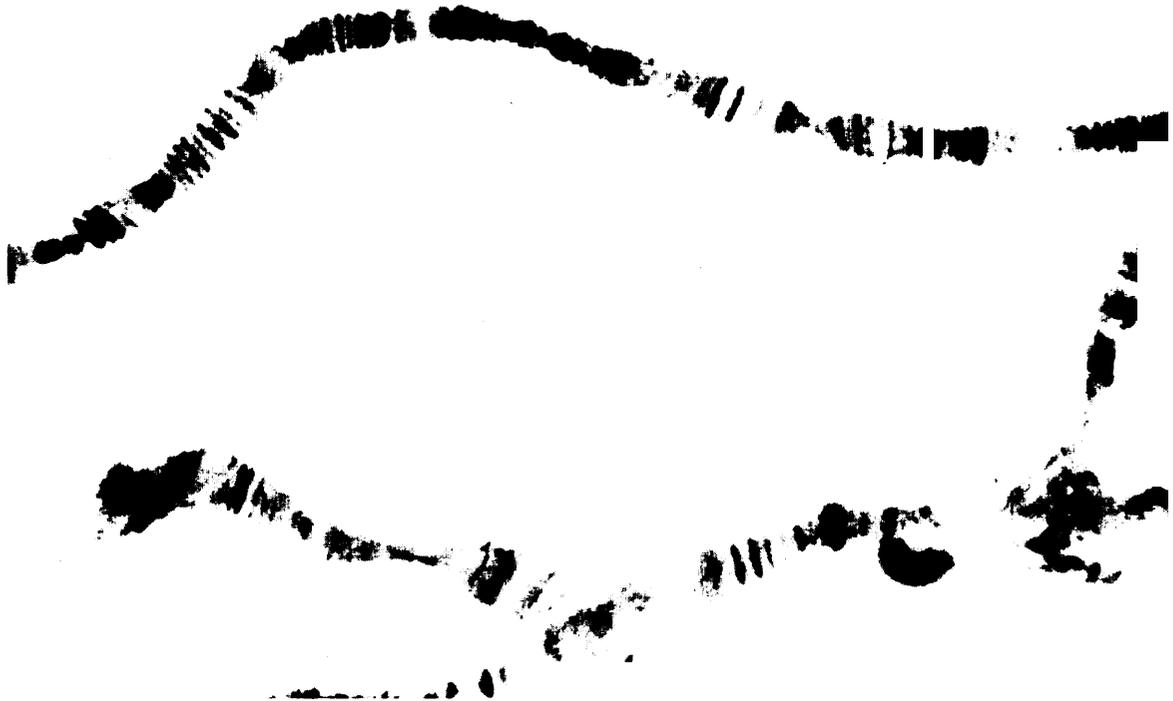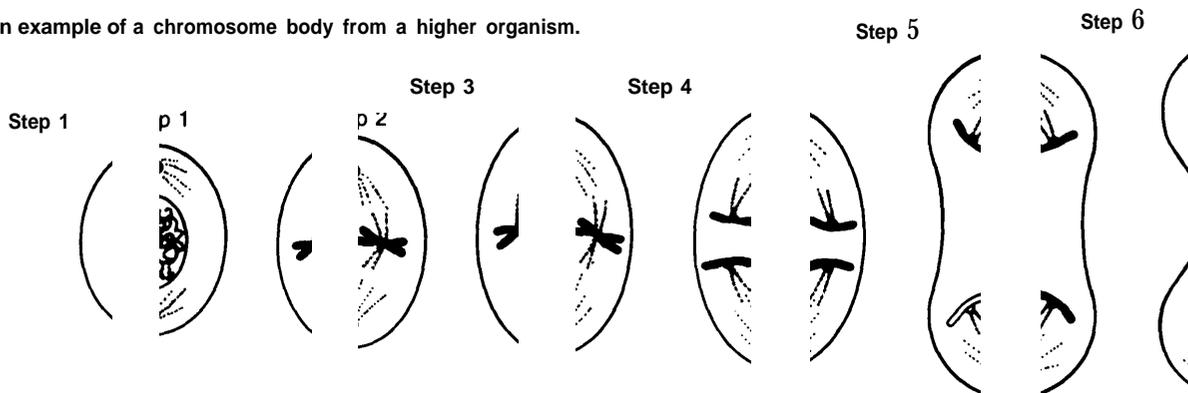
Figure 6.— Chromosomes

*Photo credit: Professor Judith Lengyel, Molecular Biology institute, **UCLA***

**Optical micrograph of chromosomal material from the salivary gland of the larva of the
common fruit fly,** *Drosophila melanogaster*

6a. An example of a chromosome body from a higher organism.

Step 1   **p 1**   **Step 3**   **Step 4**   **Step 5**   **Step 6**
              p 2

6b. In Step 1, the chromosome bodies are still uncondensed.

In Steps 2 and 3, **the chromosomes condense into thread-like bodies and align themselves near the center of the cell.**

In Steps 4 and 5, the chromosomes begin to separate and are pulled to the opposite poles of the cell.

In Step 6, the chromosomes return to an uncondensed state and the cell begins to constrict about the middle to form two new cells.

SOURCE: Office of Technology Assessment.

(kinetics in the 20th century

During the first few decades of the 20th century, scientists searched for progressively simpler experimental organisms to clarify progressively more complex genetic concepts. First was Thomas Hunt Morgan's Drosophila—gnat-sized fruit flies with bulbous eyes. These insects have a simple array of four easily distinguishable chromosome pairs per cell. They reproduce rapidly and in large numbers under the simplest of laboratory conditions, supplying a new generation every month or so. Thus, researchers could carry out an enormous number of crosses employing a whole catalog of different fruit fly traits in a relatively brief time.

It became obvious from the extensive ***Drosophila*** data that certain traits were more likely to be inherited together than others. Yellow bodies and ruby eyes, for instance, almost always went together, with both in turn, appearing more frequently than expected with the trait known as "forked bristles. " All three traits, however, showed up only randomly with curved wings. Certain genes thus seemed to be linked to one another. The entire ***Drosophila*** genome, in fact, fell into four distinct linkage groups. The physical basis for these groups, not surprisingly, consisted of the four fruit fly chromosomes. Linked genes behaved as they did because they were located on the same chromosome.

Soon, scientists learned that they could not only assign particular genes to particular ***Drosophila*** chromosomes but could identify the relative locations of different genes on a given chromosome. This gene mapping was possible because linkage itself was not permanent, linked genes sometimes separated. For instance, while yellow bodies, ruby eyes, and forked bristles were all linked traits, the first two stayed together far more frequently than either did with the third.

The degree of linkage between two genes was hypothesized to be directly proportional to the distance between them on the chromosome, mainly because of a unique event that occurs during the development of germ cells. Before the normal chromosome number is halved, the chromosomes crowd together in the center of the cell, coiling tightly around each other, practically fusing along their entire length. It is in this state that crossing-over (or natural recombination)—the actual physical exchange of parts between chromosomes—occurs. No chromosome emerges from the exchange in the same condition as before; the lengths of chromosomes are reshuffled before being transferred to the next generation.

The idea of linkage meant that Mendel's formulations had to be modified. Clearly, genes were not completely independent units. Further work with ***Drosophila*** in the 1920's showed that genes were also not permanent and could change over time. Although natural mutations occurred at a very slow rate, exposing fruit flies to X-rays accelerated their frequency enormously. Exposure of a parental fly population led to an array of new traits among their offspring—traits which, if they were neither lethal nor sterilizing, could be passed from one generation to the next.

The riddle of the gene _

With all this research, nobody yet knew what the gene was made of. The first evidence that it consisted of deoxyribonucleic acid (DNA) emerged from the work of Oswald Avery, Colin MacLeod, and Maclyn McCarty at the Rockefeller Institute in New York in the early 1940's. Avery's group took as its starting point some intriguing observations made a decade earlier by a British physician, Fred Griffith. He had worked with two types of pneumococcus (the bacteria responsible for pneumonia) and with two different bacteria within each type. One bacterium in each type was coated in a polysaccharide capsule; the other was bare. Bare bac-

teria gave rise only to bare progeny, while those with capsules produced only encapsulated forms. Only the encapsulated forms of both types II and III could cause disease; bare bacteria were benign. (See figure 7a.) But when Griffith took some encapsulated type III bacteria that had been killed and rendered harmless and mixed them with bare bacteria of type II, the presumably safe mixture became virulent: Mice injected with it died of a massive pneumonia infection, Bacteria recovered from these animals were found to be of type 11—the only living bacteria the mice had received—now wrapped in type III capsules. (See figure 7b.)

Avery's group recognized Griffith's finding as a genetic phenomenon; the dead type 111 bacteria must have delivered the gene for making capsules into the genetic complement of the living type 11 recipients. By meticulous research, Avery's group found that the substance which caused the genetic transformation was DNA.

It had been in **1868,** just 3 years after Mendel had published his findings, that DNA was discovered by Friedrich Miescher. It is an extremely simple molecule composed of a small sugar molecule, a phosphate group (a phosphorous atom surrounded by four oxygen atoms), and four kinds of simple organic chemicals known as nitrogenous (nitrogen-containing) bases. Together, one sugar, one phosphate, and one base form a nucleotide—the basic structural unit of the large DNA molecule. Because it is so simple, DNA had appeared to be little more than a monotonous conglomeration of simple nucleotides to scientists in the early 20th century. It seemed unlikely that such a prosaic molecule could direct the appearance of genetic traits while faithfully reproducing itself so that information could be transferred between generations. Although Avery's results seemed clear enough, many were reluctant to accept them.

Those doubts were finally laid to rest in a brief report published in 1953 by James Watson and Francis Crick. By using X-ray crystallographic techniques and building complex models—and without ever having actually seen the molecule itself—Watson and Crick reported that they had discovered a consistent scientifically sound structure for DNA.
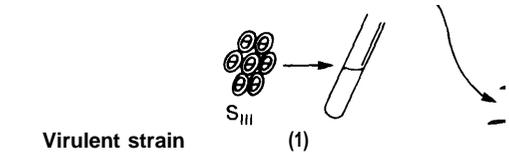
Figure 7.—The Griffith Experiment

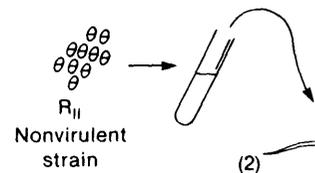**7a. There are two types of pneumococcus, each of which can exist in two forms:**

Type II

R_{II} S_{II}
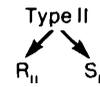
where R represents the rough, nonencapsulated, benign form; and

   S represents the smooth, encapsulated, virulent form.

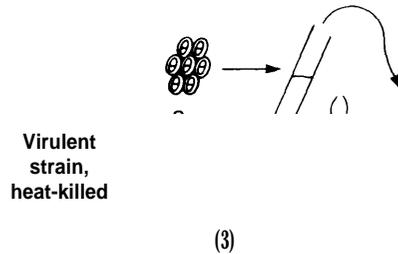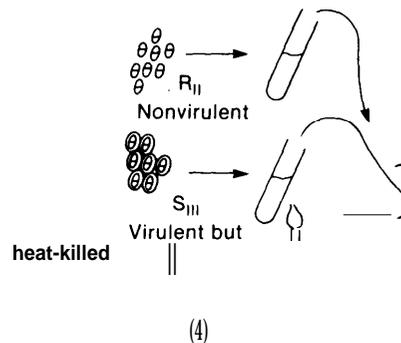**7b. The experiment consists of four steps:**

S_{III}

Virulent strain   (1)

:in𝑔   Mice injected with the vi

R_{II}

Nonvirulent strain   (2)

Mice injected with nonvirulent R_{II} do not become infected.

Virulent strain, heat-killed

(3)

The virulent S_{III} is heat-killed. Mice injected with it do not die.

R_{II}
Nonvirulent

S_{III}
Virulent but
heat-killed

(4)

When mice are injected with the nonvirulent R_{II} and the heat-killed S_{III}, they die. Type II bacteria wrapped in type III capsules are recovered from these mice.

SOURCE: Office of Technology Assessment.

The structure that Crick and Watson uncovered solved part of the genetic puzzle. According to them, the phosphates and sugars formed two long chains, or backbones, with one nitrogenous base attached to each sugar. The two backbones were held together like the supports of a ladder by weak attractions between the bases protruding from the sugar molecules. Of the four different nitrogenous bases—adenine, thymine, guanine, and cytosine—attractions existed only between adenine(A) and thymine(T), and between guanine(G) and cytosine(C). (See figure 8a) Thus, if a stretch of nucleotides on one backbone ran:

ı·Λ. . . .

the other backbone had to contain the directly opposite complementary sequence:

T-A-G-A-A-T-T. . .

The complementary pairing between bases running down the center of the long molecule was responsible for holding together the two otherwise independent chains. (See figure 8b.) Thus, the DNA molecule was rather like a zipper, with the bases as the teeth and the sugar-phosphate chains as the strands of cloth to which each zipper half was sewn. Crick and Watson also found that in the presence of water, the two poly - nucleotide chains did not stretch out to full length, but twisted around each other, forming what has undoubtedly become the most glorified structure **in the history of biology-the** double helix. (See figure **8c.)**

The structure was scientifically elegant. But it was received enthusiastically also because it implied how DNA worked. As Crick and Watson themselves noted:
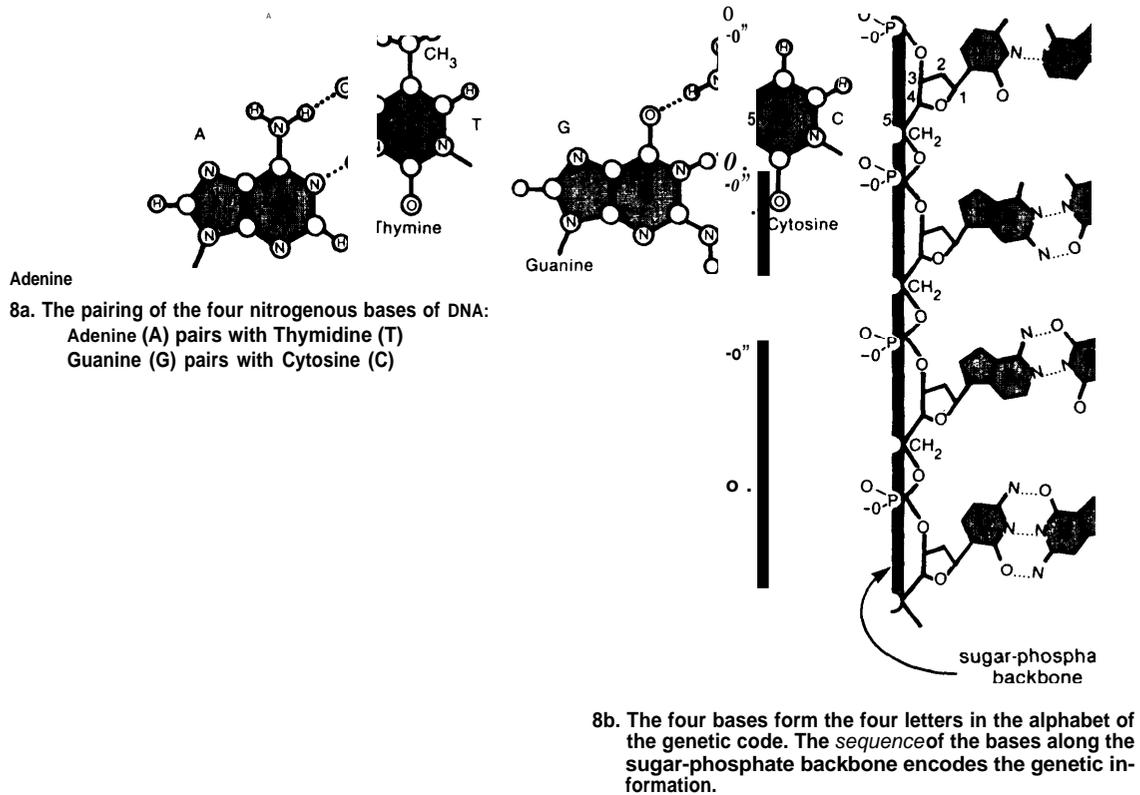
If the actual order of the bases on one of the pair of chains were given, one could write down the exact order of the bases on the other one, because of the specific pairing. Thus one chain is, as it were, the complement of the other, and it is this feature which suggests how the desoxyribonucleic acid molecule might duplicate itself. '

When a double-stranded DNA molecule is unzipped, it consists of two separate nucleotide chains, each with a long stretch of unpaired bases. In the presence of a mixture of nucleotides, each base attracts its complementary match in accordance with the inherent affinities of adenine for thymine, thymine for adenine, guanine for cytosine, and cytosine for guanine. The result of this replication is two DNA molecules, both precisely identical to each other and to the original molecule—which explains the faithful duplication of the gene for passage from one generation to the next. (See figure 9.)
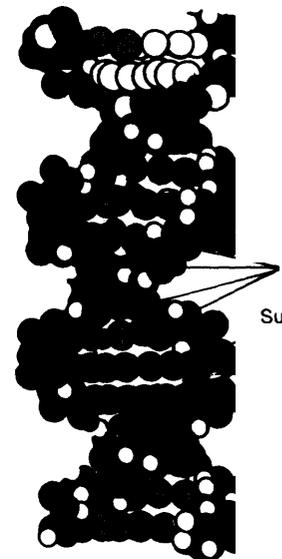
Crick and Watson's work solved a major riddle in genetic research. Because George Beadle and Edward Tat urn had recently discovered that genes control the appearance of specific proteins, and that one gene is responsible for producing one specific protein, scientists now knew what the genetic material was, how it replicated, and what it produced. But they had yet to determine how genes expressed themselves and produced proteins.

son an **D. Watson and Francis Crick,** ılications с **Implications of the Structures of Deoxyribose Nucleic Acid,** ” 53. pp. **171, 1953. pp. 737-8.**

Figure 8.—The Structure of DNA

8a. The pairing of the four nitrogenous bases of DNA:
Adenine (A) pairs with Thymidine (T)
Guanine (G) pairs with Cytosine (C)

sugar-phospha
backbone

8b. The four bases form the four letters in the alphabet of the genetic code. The *sequence* of the bases along the sugar-phosphate backbone encodes the genetic information.

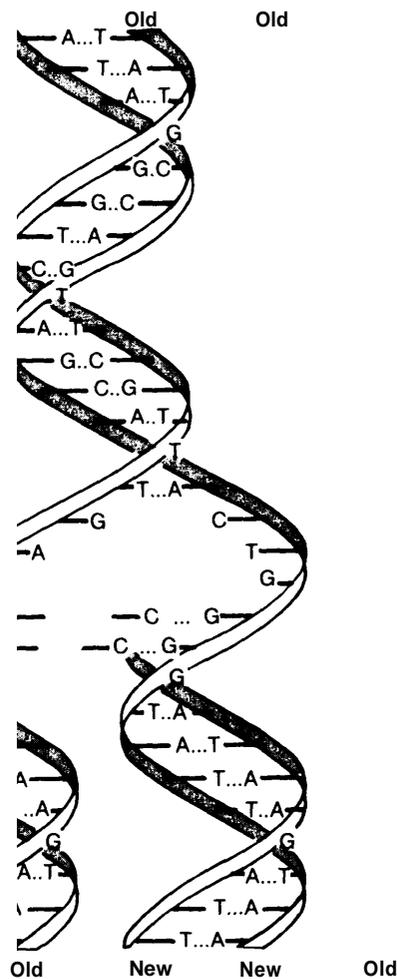A schematic diagram of the DNA double helix.   A three-dimensional representation of the DNA double helix.

&. The DNA molecule is a double helix composed of two chains. The sugar-phosphate backbones twist around the outside, with the paired bases on the inside serving to hold the chains together.

SOURCE: Office of Technology Assessment.

Figure 9.—Replication of DNA

When DNA replicates, the original strands unwind and
serve as templates for the building of new complementary
strands. The daughter molecules are exact copies of the
parent, with each having one of the parent strands.

SOURCE: Office of Technology Assessment.

The genetic code

Proteins are the basic materials of cells. Some
proteins are enzymes, which catalyze reactions
within a cell. In general, for every chemical re-
action in a living organism, a specific enzyme is
required to trigger the process. Other proteins
are structural, comprising most of the raw ma-
terial that forms cells.

Ironically, proteins are far more complex and
diverse than the four nucleotides that help
create them. Proteins, too, are long chains made
up of small units strung together. In this case,
however, the units are amino acids rather than
nucleotides—and there are **20** different kinds of
amino acids. Since an average protein is a few

hundred amino acids in length, and since any one of 20 amino acids can fill each slot, the number of possible proteins is enormous. Nevertheless, each protein requires the strictest ordering of amino acids in its structure. Changing a single amino acid in the entire sequence can drastically change the protein's character.
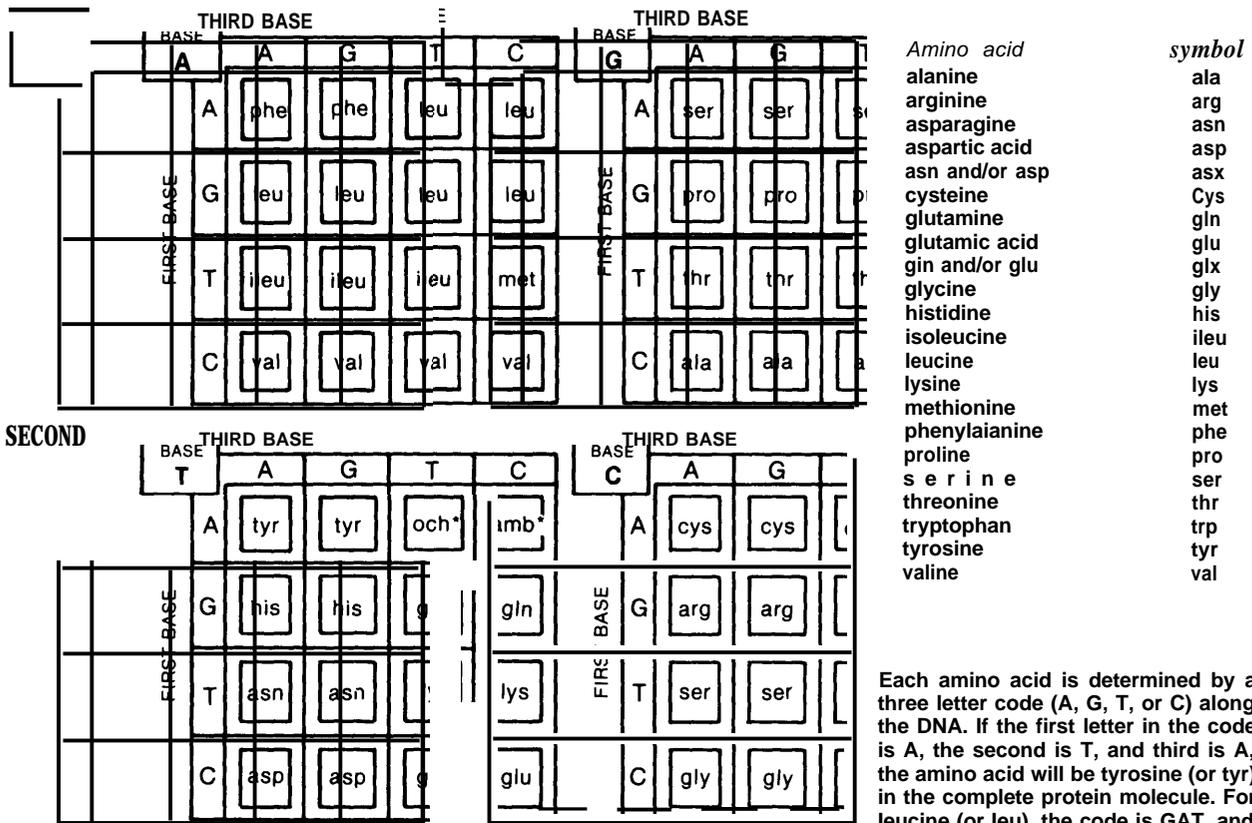
It was now possible for scientists to move nearer to an appreciation of how genes functioned. First had come the recognition that DNA determined protein; now it was evident that the sequence of nucleotides in DNA determined a linear sequence of amino acids in proteins.

By the early 1980's, the way proteins were manufactured, how their synthesis was regulated, and the role of DNA in both processes were understood in considerable detail. The process of transcribing DNA's message-carrying the message to the cell's miniature protein factories and building proteins-took place through a complex set of reactions. Each amino acid in the protein chain was represented by three nucleotides from the DNA. That three-base unit acted as a word in a DNA sentence that spelled out each protein-the genetic code. (See figure 10.)

Through the genetic code, an entire gene—a linear assemblage of nucleotides-could now be

Figure 10.—The Genetic Code

| Amino acid	symbol
alanine	ala
arginine	arg
asparagine	asn
aspartic acid	asp
asn and/or asp	asx
cysteine	Cys
glutamine	gln
glutamic acid	glu
gin and/or glu	glx
glycine	gly
histidine	his
isoleucine	ileu
leucine	leu
lysine	lys
methionine	met
phenylaianine	phe
proline	pro
s e r i n e	ser
threonine	thr
tryptophan	trp
tyrosine	tyr
valine	val

Each amino acid is determined by a three letter code (A, G, T, or C) along the DNA. If the first letter in the code is A, the second is T, and third is A, the amino acid will be tyrosine (or tyr) in the complete protein molecule. For leucine (or leu), the code is GAT, and so forth. The dictionary above gives the entire code,

● och (ochre); amb (ambert, and end are step signals for translation, i.e., signals the end of synthesis of the protein chain.
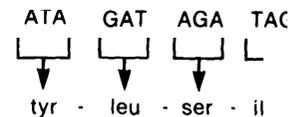
read like a book. By the *1970's,* researchers had learned to read the code of certain proteins, synthesize their **DNA,** and insert the DNA into bacteria so that the protein could be produced. (See figure 11.)

Meanwhile, other scientists **were studying** the genetics of viruses and bacteria. The combination of these studies with those investigating the genetic code led to the innovations of **genetic engineering.**

Figure 11.—The Expression of Genetic Information in the Cell
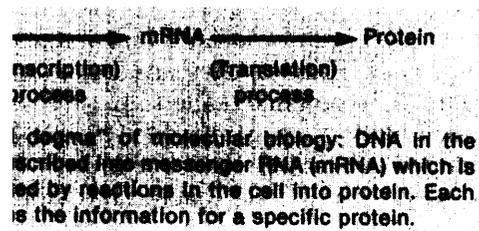
SOURCE: Office of Technology Assessment.

Developing genetic technologies

In the **early 1960's,** scientists discovered exactly how genes move from one bacterium to another. One such mechanism uses bacteriophages-viruses that infect bacteria-as intermediaries. Phages act like hypodermic needles, injecting their DNA into bacterial hosts, where it resides before being passed along from one generation to the next as part of the bacterium's own DNA. Sometimes, however, the injected DNA enters an active phase and produces a crop of new virus particles that can then burst out of their host. Often during this process, the viral DNA inadvertently takes a piece of the bacterial

DNA along with it. Thus, when the new virus particles now infect other bacteria, they bring along several genes from their previous host. This viral transduction-the transfer of genes by an intermediate viral vector or vehicle-could be used to confer new genetic traits on recipient bacteria. (See figure 12.)

Bacteria also transfer genes directly in a process called 'conjugation, in which one bacterium attaches small projections to the surface of a nearby bacterium. DNA from the donor bacterium is then passed to the recipient through the

Figure 12.—Transduction: The Transfer of Genetic Material in Bacteria by Means of Viruses
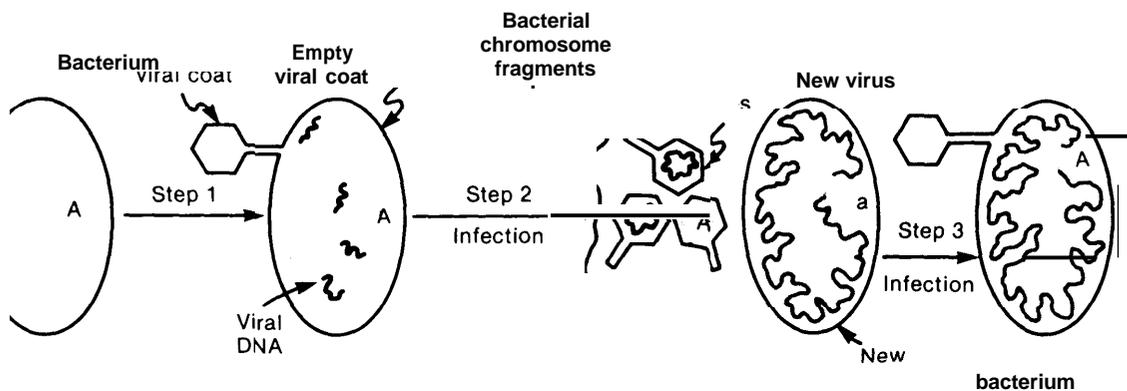
In step 1 of viral transduction, the infecting virus injects its DNA into the cell. In step 2 when the new viral particles are formed, some of the bacterial chromosomal fragments, such as gene A, may be accidently incorporated into these progeny viruses instead of the viral DNA. In step 3 when these particles infect a new cell, the genetic elements incorporated from the first bacterium can recombine with homologous segments in the second, thus exchanging gene A for gene a.

SOURCE: Office of Technology Assessment

projections. The ability to form projections and donate genes to neighbors is a genetically controlled trait. The genes controlling this trait, however, are not located on the bacterial chromosomes. Instead, they are located on separate genetic elements called plasmids-relatively small molecules of double-stranded DNA, arranged as closed circles and existing autonomously within the bacterial cytoplasm. (See figure 13.)

Plasmids and phages are two vehicles—or vectors-for carrying genes into bacteria. As such, they became tools of genetic engineering; for if a specifically selected DNA could be introduced into these vectors, it would then be possible to transfer into bacteria the blueprints for proteins—the building blocks of genetic characteristics.

But bacteria had been confronting the invasion of foreign DNA for millennia, and they had evolved protective mechanisms that preserved their own DNA while destroying the DNA that did not belong. Bacteria survive by producing restriction enzymes. These cut DNA molecules in places where specific sequences of nucleotides occur—snipping the foreign DNA, yet leaving the bacteria's own genetic complement alone. The first restriction enzyme that was iso-

lated, for instance, would cut DNA only when it located the sequence:

$$(G-A-A-)$$
$$(C-T-T)$$

If the sequence occurred once in a circular plasmid, the effect would simply be to open the circle. If the sequence were repeated several times along a length of DNA, the DNA would be chopped into several small pieces.

By the late 1970's, scores of different restriction enzymes had been isolated from a variety of bacteria, with each enzyme having a unique specificity for one specific nucleotide sequence. These enzymes were another key to genetic engineering: they not only allowed plasmids to be opened up so that new DNA could be inserted, but offered a way of obtaining manageable pieces of new DNA as well. (See figure 14.) Using restriction enzymes, almost any DNA molecule could be snipped, shaped, and trimmed with precision.

Cloning DNA—that is obtaining a large quantity of exact copies of any chosen DNA molecule by inserting it into a host bacterium-became technically almost simple. The piece in question was merely snipped from the original molecule, inserted into the vector DNA, and provided with

Figure 13.—Conjugation: The Transfer of Genetic Material in Bacteria by Mating
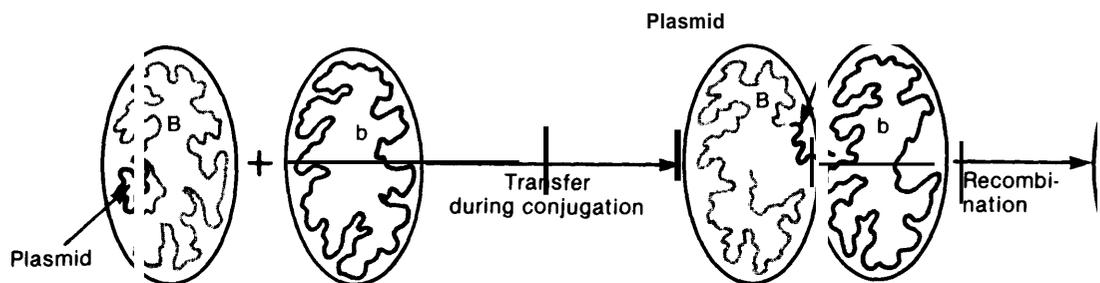
In conjugation, a plasmid inhabiting a bacterium can transfer the bacterial chromosome to a second cell where homologous segments of DNA can recombine, thus exchanging gene B from the first bacterium for gene b from the second.

SOURCE: Office of Technology Assessment.

Figure 14.—Recombinant DNA: The Technique of Recombining Genes From One Species With Those From Another
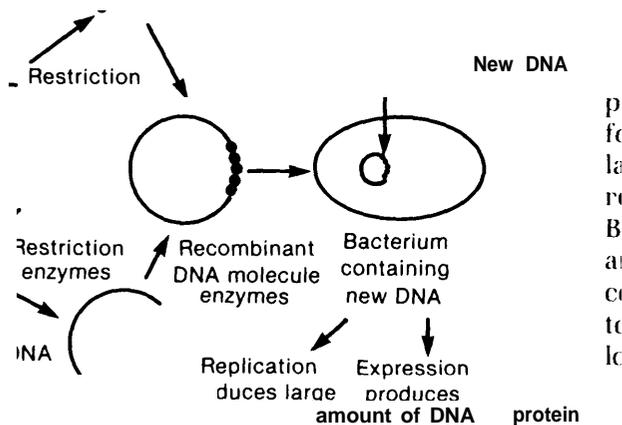
Restriction enzymes recognize certain sites along the DNA and can chemically cut the DNA at those sites. This makes it possible to remove selected genes from donor DNA molecules and insert them into plasmid DNA molecules to form the recombinant DNA. This recombinant DNA can then be cloned in its bacterial host and large amounts of a desired protein can be produced.

SOURCE: Office of Technology Assessment.

a bacterial host as a suitable environment for replication. The desired piece of DNA could be recombined with a plasmid vector, a procedure that gave rise to recombinant DNA (rDNA), also known as gene splicing. Since bacteria can be grown in vast quantities, this process could result in large-scale production of otherwise scarce and expensive proteins.

Although placing genes inside of bacteria is now a relatively straightforward procedure, obtaining precisely the right gene can be difficult. Three techniques are currently available:

● Ribonucleic acid—RNA—is the vehicle through which the message of DNA is read and transcribed to form proteins. The RNA that carries the message for the desired protein is first isolated. An enzyme, called 'reverse transcriptase, is then added to the RNA. The enzyme triggers the formation of DNA—reversing the normal process of protein production. The DNA is then inserted into an appropriate vector. This was the procedure used to obtain the gene for human insulin in 1979. (See figure 15.)

● The gene can also be synthesized, or created, directly, since the nucleotide sequence of the gene can be deduced from the amino acid sequence of its protein product. This procedure has worked well for small proteins—like the growth regulatory hormone somatostatin—which have relatively short stretches of DNA coding. But somatostatin is a tiny protein, only 14 amino acids long. With three nucleotides coding for each amino acid, scientists had to synthesize a DNA chain 42 nucleotides long to produce the complete hormone. For larger proteins, the gene-synthesis approach rapidly becomes highly impractical.

● The third method is also the most controversial. In this "shotgun" approach, the entire genetic complement of a cell is chopped up by restriction enzymes. Each of the DNA fragments is attached next to vectors and transferred into a bacterium; the bacteria are then screened to find those making the desired product. Screening thousands of bacterial cultures was part of the technique that enabled the isolation of the human interferon gene. *

At present, these techniques of recombination work mainly with simple micro-organisms. Scientists have only recently learned how to introduce novel genetic material into cells of higher plants and animals. These higher cells are being 'engineered' in totally different ways, by growing plant or animal cells in 'tissue culture' systems, in vitro.

Tissue culture systems work with isolated cells, with entire pieces of tissue, and to a far more limited extent, with whole organs or even early embryos. The techniques make it possible to manipulate cells experimentally and under controlled conditions. Several techniques are available. For example, in one set of experiments, complete plants have been grown from single cells—a breakthrough that may permit

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"Strictly speaking, RNA was transcribed using the shotgun approach into DNA, which was then cloned into bacteria and screened.

**Figure 15.-An Example of How the Recombinant DNA Technique May Be Used
To Insert New Genes Into Bacterial Cells**

The first part of the technique involves the manipula-
tions necessary to isolate and reconstruct the desired
gene from the donor:

a) **The RNA that carries the message (mRNA) for the
desired protein product is isolated.**

b) **The double-stranded DNA is reconstructed from the
mRNA.**

c) **in the final step of this sequence, the enzyme ter-
minal transferase acts to extend the ends of the
DNA strands with short sequences of identical
bases (in this case four guanines).**

I

**Messenger RNA
from animal cell**

a)

**Enzymatic
reconstruction**

Double-strand DNA   t

b)

**Terminal
transferase**

c)
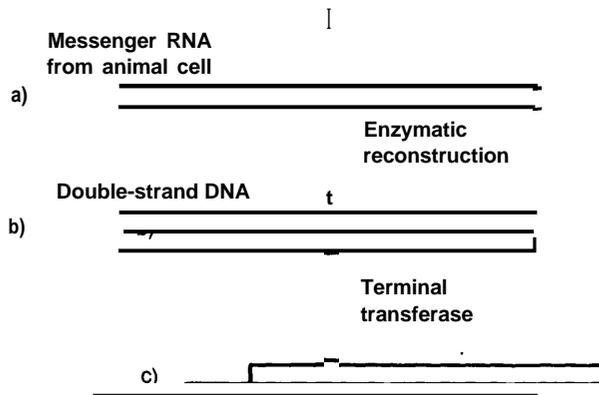
III. **The final product, a bacterial plasmid containing the
new gene, is obtained. This plasmid can then be in-
serted into a bacterium where it can be replicated and
produce the desired protein product:**

a) **The gene obtained in part i and the plasmid DNA
from part ii are mixed together and ether
because of the complementary base-pairing be-
tween them.**

b) **Bacterial enzymes fill in any gaps in the circle,
sealing the connection between the plasmid DNA
and the inserted DNA to generate an intact cir-
cular plasmid now containing a new gene.**
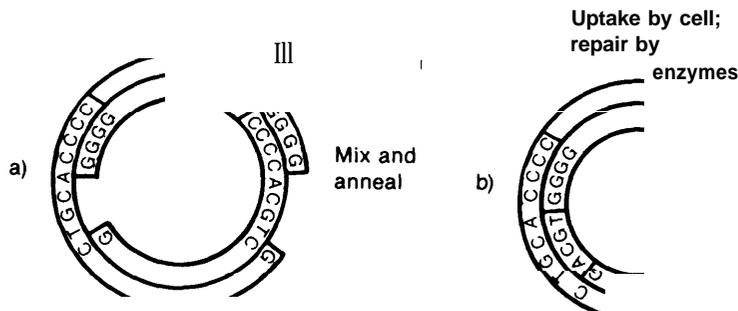
11. **A bacterial plasmid, which is a small piece of circular
DNA, serves as the vehicle for introducing the new gene
(obtained in part I above) into the bacterium:**

a) **The circular plasmid is cleaved by the appropriate
restriction enzyme.**

b) **The enzyme terminal transerase extends the DNA
strands of the broken circle with identical bases
(four cytosines in this case, to allow *complemen-
tary base pairing* with the guanines added to the
gene obtained in part i).**

II

Bacterial plasmid DNA

a)

III

a)   **Mix and
anneal**   b)

**Uptake by cell;
repair by
enzymes**
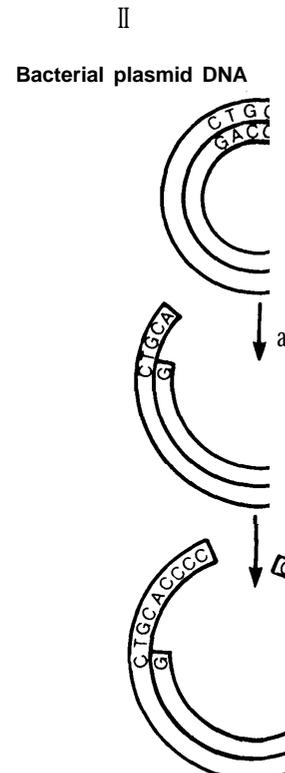
hundreds of plants to be grown asexually from a small sample of plant material. Just as with bacteria, the cells can be induced to take up pieces of DNA in a process call transformation. They can also be exposed to mutation-causing agents so that they produce mutants with desired properties. in another set of experiments, two different cells have been fused to form a new, single-cell "hybrid" that contains the genetic complements of both antecedents. In both cases, the success of tissue culture and cell

fusion* can be used to direct efficient, fast genetic changes in plants. (See ch. 8.)

Cell culture techniques, while not strictly genetic manipulation, form a major aspect of modern biotechnology. Combined with genetic approaches, their potential is only on the verge of being realized.

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● A related technique is protoplasm fusion, or the fusion of cells whose walls have been removed to leave only membrane-bound cells. The cells of bacteria, fungi, and plants must all be freed of their walls before they can be fused.

The basic issues

Applied genetics is like no other technology. By itself, it may enable tremendous advances in conquering diseases, increasing food production, producing new and cheaper industrial substances, cleaning up pollution, and understanding the fundamental processes of life. Because the technology is so powerful, and because it involves the basic roots of life itself, it carries with it potential hazards, some of which might arise from basic research, others of which may stem from its applications.

As the impacts of genetic technologies are discussed, two fundamental questions must be kept in mind:

How will applied genetics be used?

Interest in the industrial use of biological processes stems from a merging of two paths: the revolution in scientific understanding of the nature of genetics; and the accelerated search for a sustainable society in which most industrial processes are based on the use of renewable resources. The new genetic technologies will spur that search in three ways: they will provide a means of doing something biologically—with renewable raw materials—that previously required chemical processes using nonrenewable resources; they will offer more efficient, more economical, less polluting ways for producing both old and new products; and they will increase the yield of the plant and animal resources that are responsible for providing the world's supplies of food, fibers, and some fuels.

What are the dangers?

Even before scientists recognized the potential power of applied genetics, some questioned its consequences; for with its benefits, appeared hypothetical risks. Although most experts today agree that the immediate hazards of the basic research itself appear to be minimal, nobody can be certain about all the consequences of placing genetic characteristics in micro-organisms, plants, and animals that have never carried them before. There are at least three separate areas of concern:

First, genetically engineered micro-organisms might have potentially deleterious effects on human health, other living organisms, or the environment in general. Unlike toxic chemicals, organisms may reproduce and spread of their own accord; if they are released into the envi -ronment, they may be impossible to control.

Second, some observers question whether sufficient knowledge exists to allow the extinction of diverse species of "genetically inferior" plants and animals in favor of a few strains of "superior" ones. Evolution thus far has depended, in part, on genetic diversity; replacing in nature diverse inferior strains by genetically engineered superior strains may increase the susceptibility of living things to disease and environmental insults.

Finally, this new knowledge affects the understanding of life itself. It is tied to the ultimate

questions of how humans view themselves and what they legitimately control in the world.

Because of the significant and wide-ranging scope of applied genetics, society as whole must begin to debate the issues with a view toward allocating and monitoring its benefits and burdens. That process requires knowledge. The following sections of the report describe the impacts of applied genetics on specific industries, and assess many of their consequences.