

HC70A

Winter 2004

Professor Bob Goldberg

Lecture #1

The Age of DNA -

What is Genetic Engineering?

Themes/Concepts

1 2.5 hr  
Lecture 1  
15 min Lecture 2

- ✓ ① Age of DNA & Genetic Engineering *DNA Splicing*
- ✓ ② DNA → RNA → Protein Universal & Allows Genetic Engineering
- ✓ ③ Why Clone Genes?
- ✓ ④ How has DNA Manipulation Affected Society?
- ✓ ⑤ DNA Manipulation has lead to the Era of Genomics
- ✓ ⑥ What is genetic Engineering? Anything New?
- ✓ ⑦ Classical vs. Molecular Genetic Engineering *vegetables eugenics DNA*
- ✓ ⑧ ~~STOP~~ Genetic Engineering *1/8/04* & ~~Age of~~ *cut* ~~Reproduction!~~ *Lecture #2*
- ✗ ⑨ Examples of Genetic Engineering
- ✓ ⑩ What is HC70A about & who are the Players? *1/8/04*

An Age of DNA that  
comes into the home....



**MOLECULAR GENEALOGY RESEARCH PROJECT** Uses mouthwash. FREE.  
[molecular-genealogy.byu.edu](http://molecular-genealogy.byu.edu)

**FAMILY TREE DNA** No blood—just swab your cheek. \$149 AND UP.  
[dnafamilytree.com](http://dnafamilytree.com)

**SCIENCE**

## DO-IT-YOURSELF DNA

If you've tried and tried but your family tree is still just a seedling, mail-order DNA testing may be for you. Comparing your genetic profile with those of other genealogy buffs—and potential relatives—can provide new leads. For \$149 and up, Family Tree DNA will give you a list of 25 markers (or genetic traits) you carry, based on a swab from the inside of your cheek. For a bit more—\$220 and up—Oxford Ancestors ([oxfordancestors.com](http://oxfordancestors.com)) will check 10 markers and tell you which "Seven Daughters of Eve" clan you belong to. If that's too steep, the Molecular Genealogy Research Project will test 250 markers for free. Run by Brigham Young University, it hopes to create a worldwide database. The catch: the data must be kept anonymous. In other words, the project will create a map of ancestry lines—not an individual report for you.

—MATTHEW MACROBERTS

Do it yourself DNA testing  
to find family history!!  
\$200

YOU will have a DNA Test this Quarter!

WE LIVE in The Age of DNA

The Age of the Gene

April 4th...



**DNA**  
P E R F U M E<sup>®</sup>



by  
**bijan**

DNA...

it's not just  
a perfume...  
it's gene therapy.

we have begun to *control* our Biological Destiny!

WHAT DOES DNA LOOK & FEEL LIKE?

①



# Discover

JULY 2003

SCIENCE, TECHNOLOGY, AND MEDICINE

Now the  
**Genetic**  
Testing  
Really  
Begins

It Starts With  
a Single  
Drop  
of **Blood**  
Taken From  
Each Newborn

And Ends When  
Scientists Predict  
Everyone's  
Physical  
and Mental  
**Future**

Human red blood cells. Magnification: 19,600x





# Do you want to know your future?

Every state in the country requires that infants be tested for a list of obscure diseases. Before long, some

states could move on to DNA testing of all newborns. Now is the time to decide a critical question: How much do we want to know and when do we want to know it?

By Jeff Wheelwright

Photography by Catherine Ledner

TESTING  
YOUR  
FUTURE



DNA TESTING IS USED IN MANY  
Situations involving Identity —  
in addition to Humans

## DNA Confirms Infected Cow's Origin

Next in the inquiry  
into a Washington state  
case of mad cow disease  
is a focus on feed.

By JOHANNA NEUMAN  
Times Staff Writer

WASHINGTON — DNA tests have confirmed that the Holstein found last month to be infected with mad cow disease originated in Alberta, Canada, U.S. Department of Agriculture officials said Tuesday.

The DNA testing on the cow and her offspring, as well as earlier-reported records showing that the cow had been sold by an Alberta farmer disposing of his dairy herd, "makes us confident in the accuracy of this trace-back," said W. Ron DeHaven, the department's chief veterinarian.

The confirmation, based on DNA tests at two laboratories — one in the United States and one in Canada — leaves unanswered the question of how the cow from a farm in Washington state became infected. Officials will now concentrate on the feed used by the cow's original owner in Alberta.

Dr. Brian Evans, chief veterinary officer for the Canadian

Food Inspection Agency, said on a USDA conference call Tuesday that investigators would also try to determine whether the feed source for the Holstein was the same as that for an Alberta cow diagnosed with mad cow disease in May.

Scientists believe that bovine spongiform encephalopathy, or BSE, the brain-rotting illness commonly known as mad cow disease, can be transmitted to cattle that eat feed containing the remains of infected cows. In the past, leftover parts of slaughtered animals — including the brain and the spinal cord, which are believed to harbor the source of the infection — were ground up and used in animal feed.

In 1997, the U.S. and Canada banned the use of the remains of ruminants, or cud-chewing animals, in feed used for cattle, but both North American cows diagnosed with BSE — the one discovered in Canada in May and the one found in the United States in December — were born several months before that ban went into effect. The human form of the illness, variant Creutzfeldt-Jakob disease, has been associated with consumption of food made from BSE-infected animals.

Agriculture Secretary Ann

M. Veneman announced Dec. 23 that a cow slaughtered Dec. 9 had tested positive for BSE. The cow was tagged for testing because it was a "downer" cow that was unable to walk to slaughter. The cow's meat products had already been distributed before Veneman's announcement, primarily to retail outlets in Washington and Oregon.

While officials recalled the meat, it is not known how much was recovered.

Veneman has since announced a series of reforms to bolster U.S. defenses against BSE, including a ban on accepting downer cows for slaughter and a rule that would hold all meat products from an animal tested for disease until results are completed.

But after Tuesday's announcement of the DNA results confirming the cow's origin, some producers said the Agriculture Department had moved too slowly to determine the source of the infection.

"They knew the leads pointed back to Canada, and if they had made the announcement immediately, it might have mitigated a great deal of our loss," said John Lockie, executive director of R-CALF USA, a national association of cattle producers.

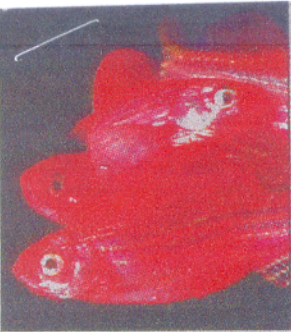


THE AGE OF GENETIC ENGINEERING  
COMES INTO THE HOME.....

Genetically Engineered Zebra fish

## State Takes Dim View of GloFish, Bans Sale

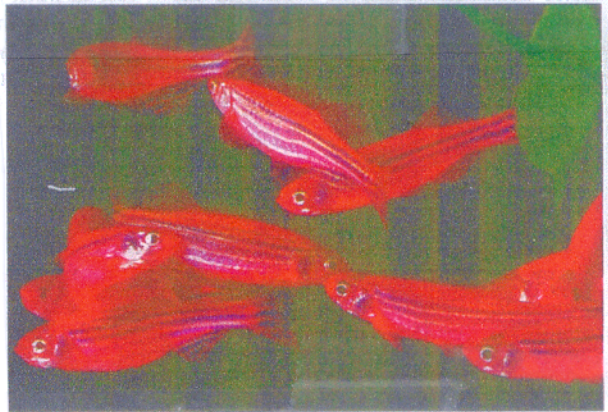
By KENNETH R. WEISS  
Times Staff Writer



PR News

**RED ZEBRA:** GloFish  
are implanted with a gene  
from sea anemones.

## State Game Panel Bans Sale of GloFish



Glowing review: watchdogs want tighter rules for transgenic pets.

# What Questions Are Generated By the GloFish?

- ① What is a Zebra Fish & How closely Related to Humans?
- ② Why Use Zebra Fish as a "Model Organism" For Studying Genes?
- ③ How was the GloFish Engineered?
- ④ What Gene Makes the GloFish Glow?!
- ⑤ How "old" is the technology used to Engineer a Transgenic GloFish?
- ⑥ What is the significance of being able to insert a "foreign" Gene in a GloFish?  
To Biology? To Genetics? To Medicine?
- ⑦ Does a pet transgenic GloFish raise regulatory issues? ethical issues? patent & commercialization issues? ecological issues?
- ⑧ What are they?



WHAT ABOUT A Glo Monkey - ANDi?

# MONKEY BUSINESS

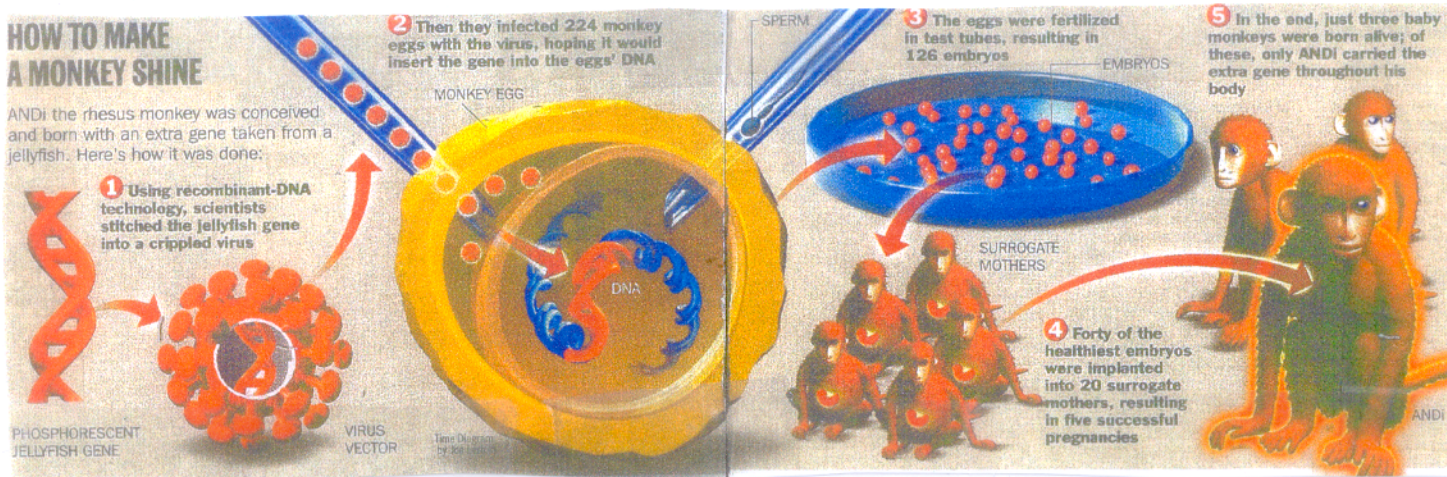
A tiny primate with a gene from a jellyfish raises scientists' hopes—and some serious ethical questions



**MADONNA AND MONKEY** Researcher Christa Martinovich cradles ANDi soon after birth; his temperament, she says, was "perfect"

## HOW TO MAKE A MONKEY SHINE

ANDi the rhesus monkey was conceived and born with an extra gene taken from a jellyfish. Here's how it was done:



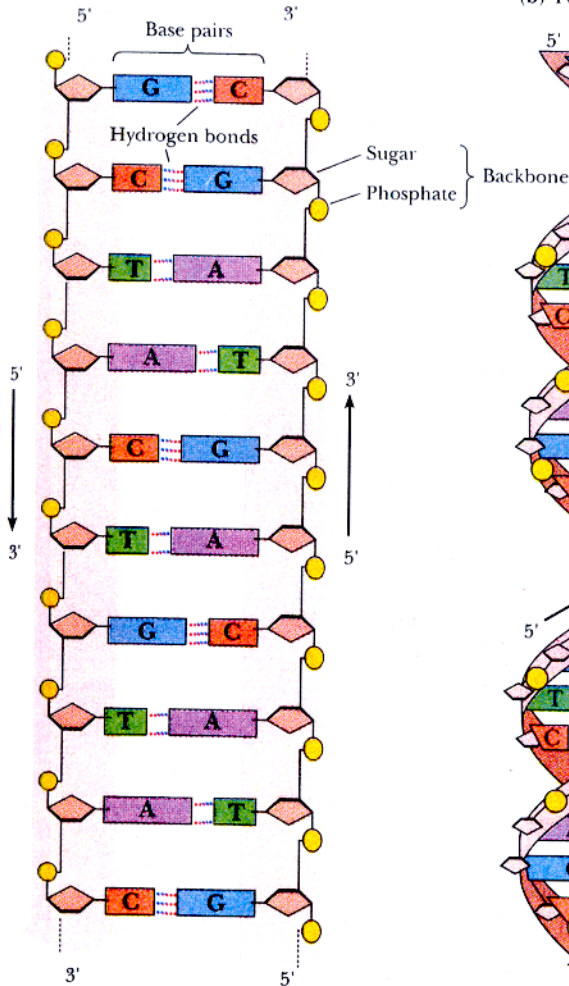
How can this technology help human beings?

Are there ethical issues in genetically engineering Monkeys? *Humans?* ← Has it been done?

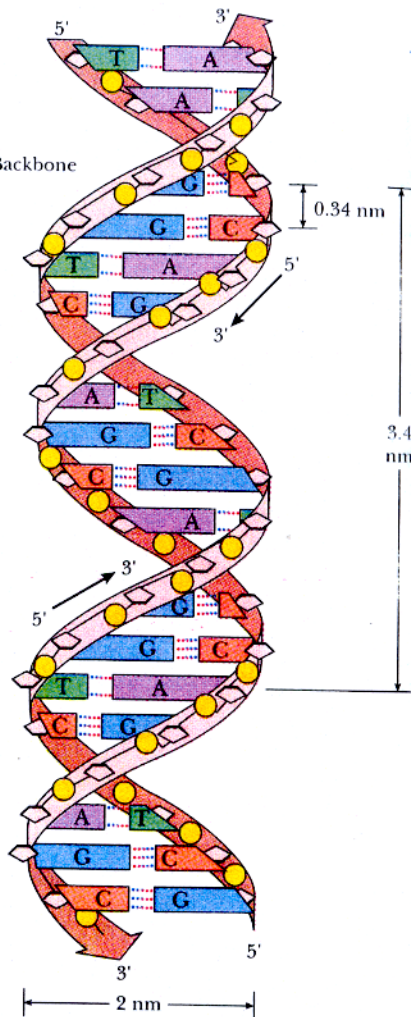


# The Chemical Structure of DNA Generates ALL of the Biological Diversity on the Earth

(a) Ladder model of DNA



(b) Twisted ladder model of DNA



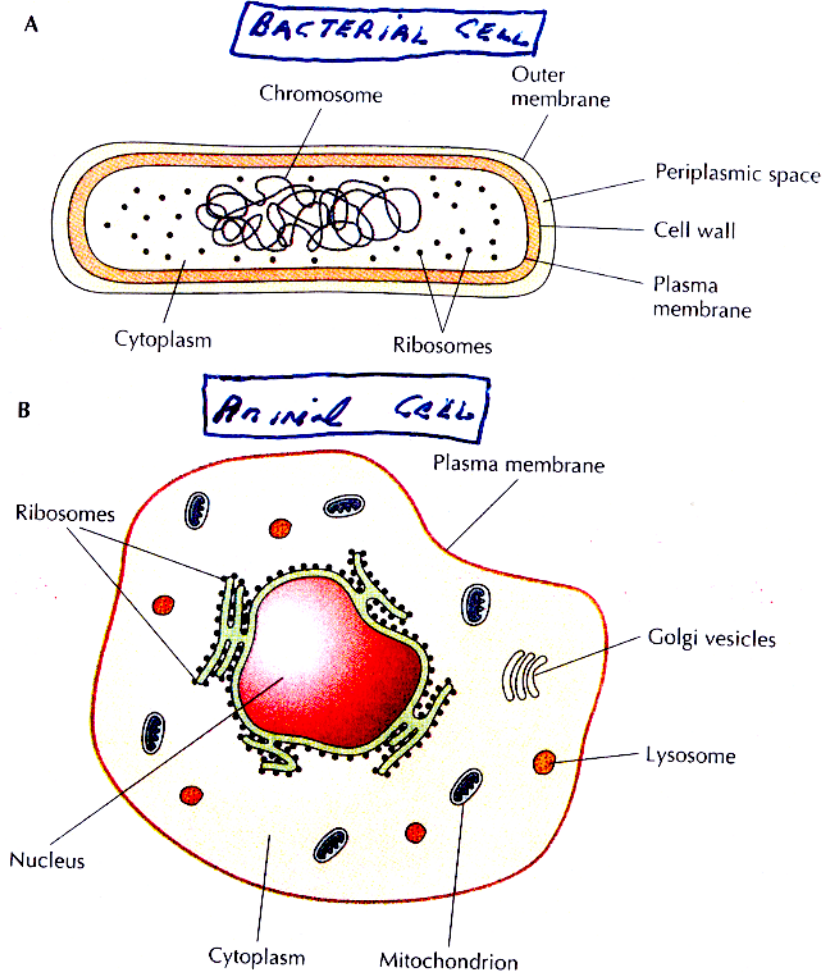
We Now Are Able to Manipulate  
the DNA of Any Organism  
using DNA Technology

NEED TO UNDERSTAND THIS TECHNOLOGY!



Cells Are Unique Because  
They Contain A Unique  
Collection of Genes!

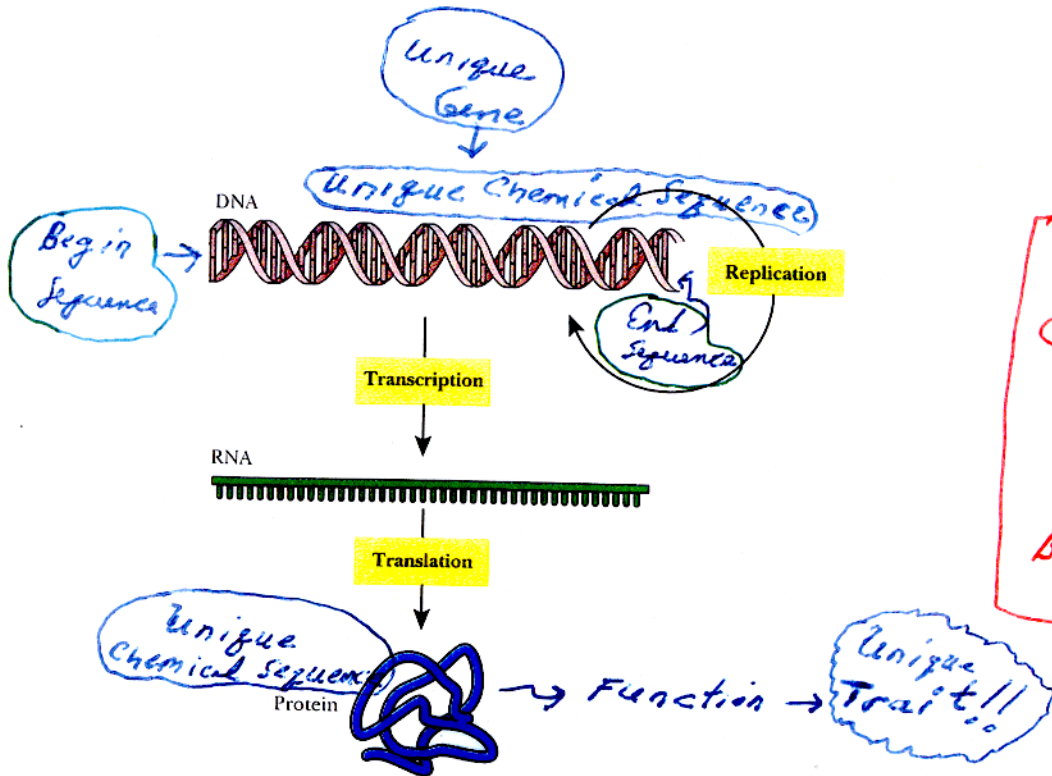
Figure 2.1 Schematic representations of a prokaryotic bacterium (A) and a eukaryotic animal cell (B).



What  
Makes  
A  
bacteria  
+  
What  
makes  
A  
Human  
??

ALL in  
the  
DNA!

Genes → Unique Biological Traits



The Central Dogma of Biology!

Genes

- ① Replicate & Pass Information to Next Generation
- ② Instruct Cells to Perform a Function  
↳ Produce a trait (e.g., eye color)

question?

CAN we use these processes to "piggy back" foreign genes?

Genes ARE Made of DNA!

These Processes Are Used For Genetic Engineering Naturally



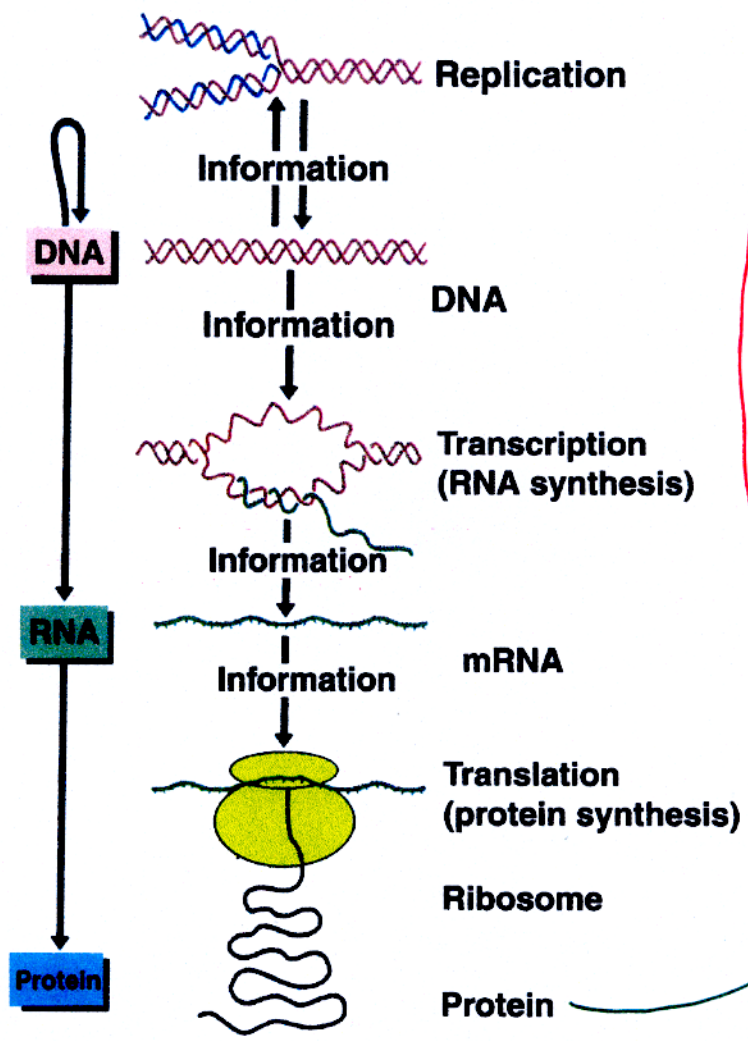
The Process of Gene to Trait  
is the "SAME" in all organisms!

UNIVERSAL PROCESS!

**Translating The Genetic  
Code Into Proteins is a  
Conserved Process**

⇒ The reason  
"why"  
genetic  
engineering  
is  
possible

CAN  
INTERVENE  
in  
this Process  
in Living  
Cells

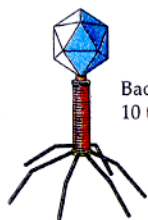


What is  
the "Big"  
Implication of  
"this" for  
Biology?

Trait  
Ceg, eye color

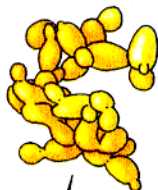
ALL ORGANISMS USE THE SAME PROCESSES  
And "RULES" to Generate Traits!!  
And the SAME MOLECULES/CHEMISTRY  
is involved!

# ORGANISMS ARE UNIQUE BECAUSE THEY HAVE A UNIQUE COLLECTION of Genes OR **Genome**



Bacteriophage (virus)  
10 thousand bp

Yeast  
27 million bp



*E. coli*  
4 million bp



We expect simple organisms to have small genomes...

*Caenorhabditis elegans* (roundworm)  
160 million bp per cell



Fruit fly  
330 million bp  
per cell



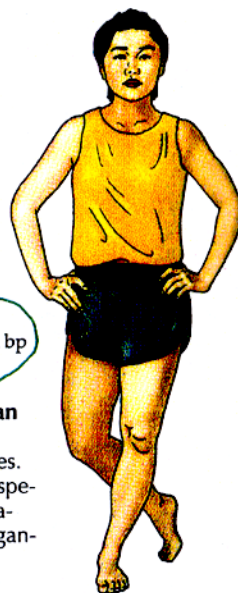
How much per sperm & egg?

Lily  
106 billion bp  
per cell



...but why does a lily have 18 times the DNA that a human does?

Human  
6 billion bp  
per cell



**14.2 Amounts of Genomic DNA Can Be Deceiving** Eukaryotes have more DNA in their genomes than prokaryotes. However, among some eukaryotes—especially plants—there is no apparent relationship between genome size and organism complexity. The question of why some plants accumulate such great amounts of seemingly nonfunctional DNA has no clear answer and continues to be investigated.

**Table 16-1 Haploid Genome Sizes (in Nucleotide Pairs) of Model Genetic Organisms**

Organism	Group	Genome Size
T4	bacteriophage	$2.0 \times 10^5$
<i>E. coli</i>	bacterium	$4.2 \times 10^6$
<i>Saccharomyces cerevisiae</i>	yeast	$1.8 \times 10^7$
<i>Neurospora crassa</i>	mold	$2.7 \times 10^7$
<i>Dictyostelium discoideum</i>	slime mold	$5.4 \times 10^7$
<i>Caenorhabditis elegans</i>	nematode	$1.0 \times 10^8$
<i>Arabidopsis thaliana</i>	plant	$1.0 \times 10^8$
<i>Drosophila melanogaster</i>	insect	$1.4 \times 10^8$
<i>Mus musculus</i>	mammal	$3.0 \times 10^9$
<i>Homo sapiens</i>	mammal	$3.3 \times 10^9$
<i>Zea mays</i>	plant	$5.4 \times 10^9$

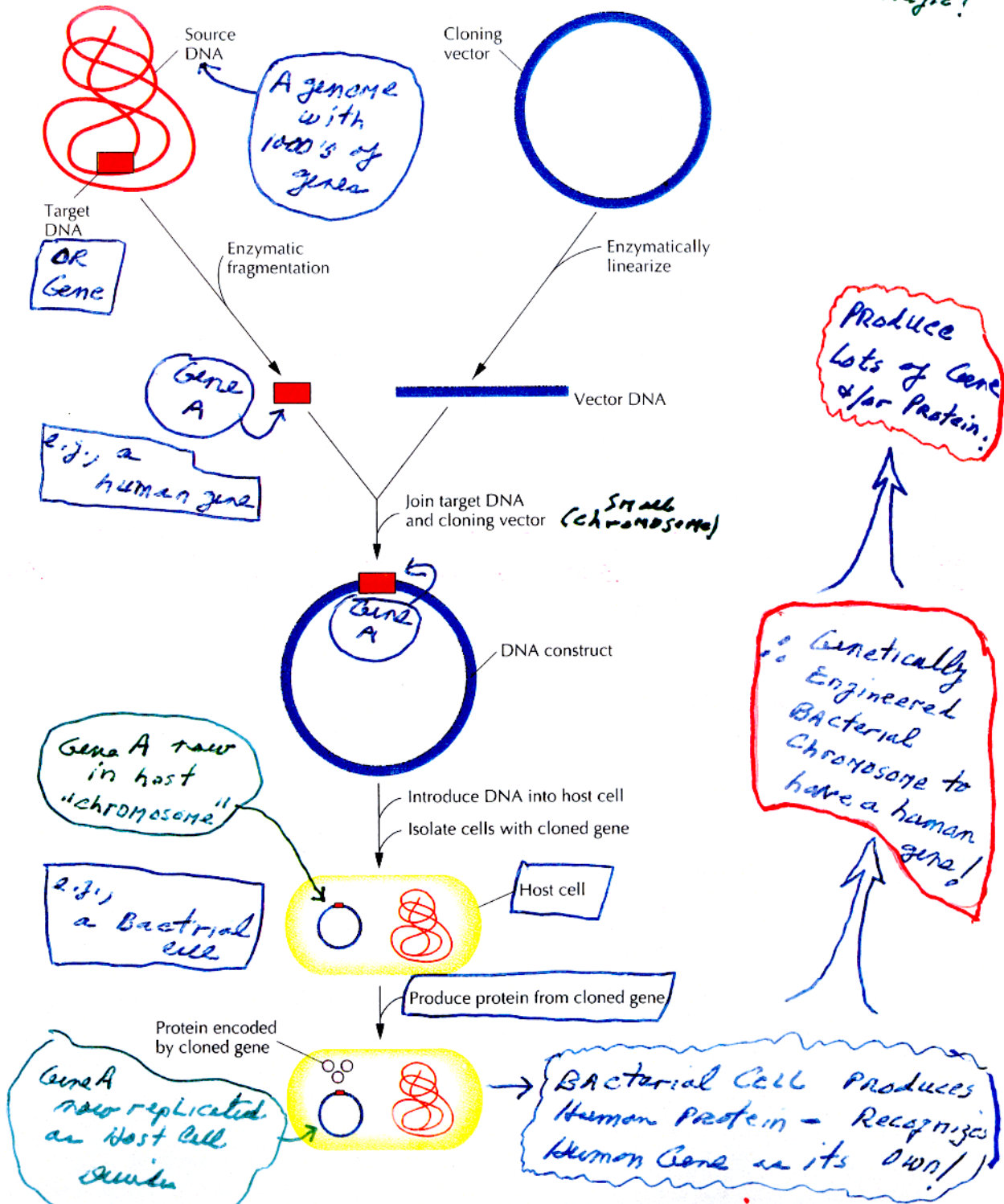
Humans have  $3 \times 10^9$  bp (3000 Mb)  
of DNA in their genome  
~1000x more than *E. coli*

Genome = Unique / Total Collection of  
Genes in an organism's chromosome(s)  
 $\therefore$  Unique Chemical Sequence of  
DNA!



DNA Cloning or Genetic Engineering  
Uses Natural Processes of living  
cells to Isolate Single Genes

NO  
"HOCUS  
POCUS"  
It's Not  
Magic!



Produce  
Lots of Gene  
& for Protein!

Genetically  
Engineered  
Bacterial  
Chromosome to  
have a human  
gene!

Implications?

# "Why" Clone Genes From the Genome of an ORGANISM?

Purify Individual Genes from the Genome --- Separate from rest of Genes

Amplify the Gene to obtain enough DNA to study and/or Engineer

Use the cloned Gene to:

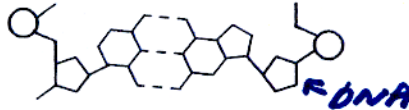
- (a) Study Gene Structure & Function
- (b) Use to make pharmaceuticals
- (c) Use in animal & plant gene therapy
- (d) Use to diagnose diseases
- (e) Use to correct diseases
- (f) Use to Identify individuals
- (g) Use to convert cells into factories

Gene Engineering has lead to new knowledge About how Living cells Function and for Applications that improve all of our Lives!



# DNA CLONING ALLOWS "US" TO ISOLATE, MANIPULATE, & Study INDIVIDUAL Genes

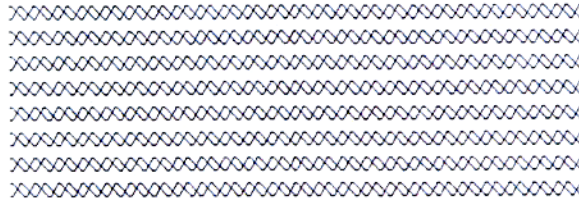
The average atomic mass of one base pair is 635 daltons (a dalton is 1/12 the mass of a carbon atom)



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The  $\beta$  globin gene is approximately 2000 bp in length  
So, the atomic mass of the  $\beta$  globin gene is:

$$\begin{aligned} &2000 \text{ bp} \\ &\times \\ &635 \text{ daltons/bp} \\ &= 1.27 \times 10^6 \text{ daltons} \end{aligned}$$

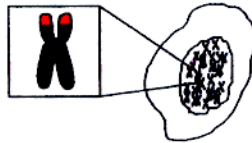


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## Mass of $\beta$ globin gene in an adult human

There are two copies of the  $\beta$  globin gene per cell



There are  $10^{13}$  cells per individual

So, the total atomic mass of  $\beta$  globin DNA per individual is:

$$\begin{aligned} &1.27 \times 10^6 \text{ daltons/gene} \\ &\times \\ &2 \text{ genes/cell} \\ &\times \\ &10^{13} \text{ cells/individual} \\ &= 2.54 \times 10^{19} \text{ daltons} \end{aligned}$$



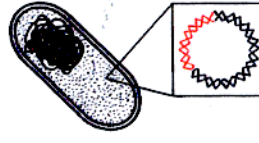
If there are  $6.02 \times 10^{23}$  daltons per gram, then:

$$\begin{aligned} &\frac{2.54 \times 10^{19} \text{ daltons}}{6.02 \times 10^{23} \text{ daltons/gram}} \\ &= 0.000042 \text{ grams} \\ &= 0.042 \text{ mg} \end{aligned}$$

0.42  $\mu$ g  $\beta$  globin DNA per human

## Mass of $\beta$ globin gene in a liter of *E. coli*

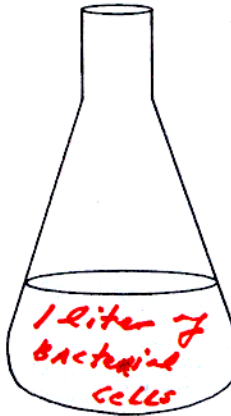
There are 500 copies of the  $\beta$  globin gene per cell



There are  $5 \times 10^{11}$  cells per liter

So, the total atomic mass of  $\beta$  globin DNA per liter is:

$$\begin{aligned} &1.27 \times 10^6 \text{ daltons/gene} \\ &\times \\ &500 \text{ genes/cell} \\ &\times \\ &5 \times 10^{11} \text{ cells/liter} \\ &= 3.175 \times 10^{20} \text{ daltons} \end{aligned}$$



If there are  $6.02 \times 10^{23}$  daltons per gram, then:

$$\begin{aligned} &\frac{3.175 \times 10^{20} \text{ daltons}}{6.02 \times 10^{23} \text{ daltons/gram}} \\ &= 0.000527 \text{ grams} \\ &= 0.527 \text{ mg} \end{aligned}$$

527  $\mu$ g  $\beta$  globin DNA per liter!

**Mass of  $\beta$  Globin DNA in Adult Human vs. 1-liter Culture of *E. coli* Carrying  $\beta$  Globin Gene on Plasmid**

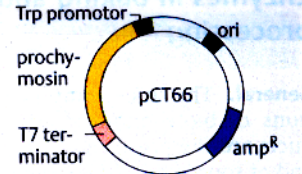
CAN Produce 10X More of a single human gene in a 1 liter bacteria culture than in the entire human body using gene engineering methods!

# Even Cheesemaking is Helped By DNA Cloning + Genetic Engineering

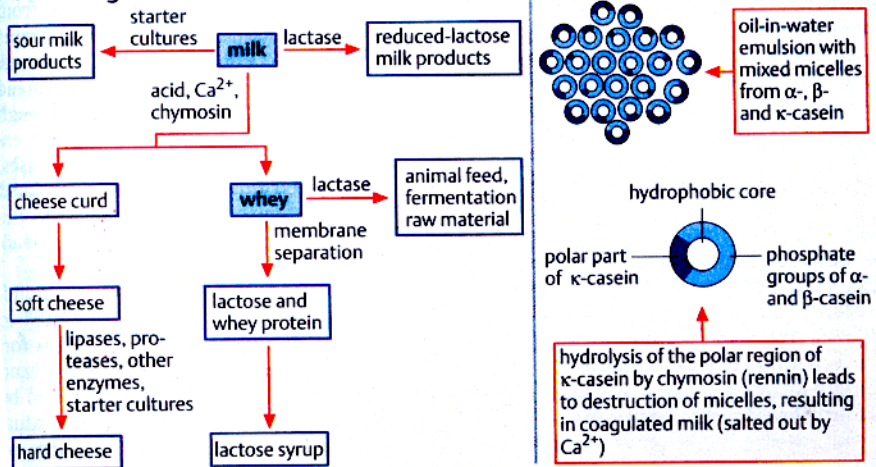
## Composition of milk

	milk (%)	whey (%)
water	~ 88	~ 94
fat	~ 3-4	~ 0.5
protein	~ 3.3	~ 1
casein	~ 2.6	-
lactose	-	~ 4.8

## Plasmid for the expression of chymosin in *E. coli*



## Processing of milk



## Manufacture of chymosin

native	microbial	recombinant
<b>stomachs of young animals</b> cutting, activation at pH < 5	<b>preculture</b> high-yield mutants of <i>Mucor miehei</i> or <i>M. pusillus</i>	<b>recombinant microorganism</b> <i>Escherichia coli</i>
<b>extraction</b> salt water, 14 d	<b>bioreactor</b> dextrose syrup, soy meal, 30°C, 72 h	<b>bioreactor</b> maltodextrins, 37°C, 36 h
<b>purification</b> ultrafiltration standardization	<b>purification</b> separation of mycelium, reverse osmosis, precipitation	<b>purification</b> isolation of inclusion bodies, Triton-X100/EDTA, urea-/alkali-extract, ion-exchange chromatography, acid treatment
200 U/kg stomach	5000 U/m <sup>3</sup> in 72 h	20000 U/m <sup>3</sup> in 36 h

100X more in a bacterial culture!

The cow chymosin gene is cloned + amplified in bacteria leading to an infinite amount of chymosin to make cheese!

What about religious issues? Kosher laws?



# The ERA OF DNA MANIPULATION MEANS.....

- ① DNA/GENES CAN BE CLONED/ISOLATED FROM ANY ORGANISM
- ② DNA segments of Any kinds and FROM any organisms CAN BE COMBINED
- ③ Engineered Gene/DNA Molecules CAN BE Re-Inserted into the cells of Any ORGANISM & Made to Work -
- ④ Whole Genomes & "organisms" can be Synthesized!

There ARE NO Genetic Limits -  
All of Biology uses the  
SAME RULES!

We Have Known How to Manipulate  
Genes FOR 25 years!

The Implications are  
ENORMOUS!

THE AGE OF DNA AND GENE  
CLONING HAS AFFECTED  
SOCIETY in MANY  
ways!

AND WE HAVE JUST  
BEGUN!

- ① Basic Understanding of Living Processes!  
what is life? what is the basis  
of Biological diversity?
- ② Basic Understanding of Genes
- ③ Medicine → Diagnosis & Treatment of Diseases
- ④ Agriculture → Higher yielding crops
- ⑤ Business / Commerce → Biotech Industry
- ⑥ The Law / Forensics
  - ↳ Patents
  - ↳ Identification
  - ↳ Privacy Issues
- ⑦ Anthropology
  - ↳ Human origins / diversity & Unity of Humankind
- ⑧ Evolution
  - ↳ Where did we come from?
- ⑨ Philosophy / Religion → How we view ourselves  
in relation to God & nature