

Bio 97

Cloning a sheep vs. cloning a gene



SCOTTISH SHEEP SHOCKER!

1996



**HELLO
DOLLY!**

Scientist in Scotland grows "impossible" sheep from one cell of another sheep.

Cloning a sheep

- Take an egg* and destroy or remove its nucleus
- Inject the enucleated egg with a new nucleus that was taken from an adult cell of the individual you want to clone
- Implant the “new embryo” into a surrogate mother

*or an embryonic stem cell

Cloning a sheep...

- Result is a genetically identical copy sheep
- Now we have two (or more) sheep with exactly the same genotype
- The only other way for that to happen is with identical twins

Gen En #46: Cloning Dolly

Ethical
Issue
of the Day #1

Reproductive Cloning

Arguments for a ban

on human reproductive* cloning

- hundreds of cloned embryos must be created and placed into a mother to produce a single live birth
- dying, stillborn and deformed babies frequent
- Even those who have lived a long time (such as Dolly) have a large number of serious health problems

*cloning to make a baby

The Boys from Brazil



Book by Ira Levin (1978)

Movie (1978)

**About an attempt to (reproductively)
clone Adolph Hitler**

What about cloning animals?

Animals reproductively cloned from adult cells:

Frog (1952)

Sheep (1996)

Mouse (1997)

Cow (1998)

Goat (2000)

Cat (2001)

Rabbit (2002)

Horse, Rat (2003)

What about cloning animals?

Genetic Savings & Clone, Inc.

www.savingsandclone.com

clone



donor



Therapeutic Cloning

**for stem cell
research has merits**

Cloning a gene

- Insert a particular fragment of DNA (a particular gene) into a vector
- A vector is another DNA molecule that can be put into a host
- This creates a new DNA molecule
--> Recombinant DNA
- Greatly aids further study of that gene, and its use in diagnosis or treatment

Vector

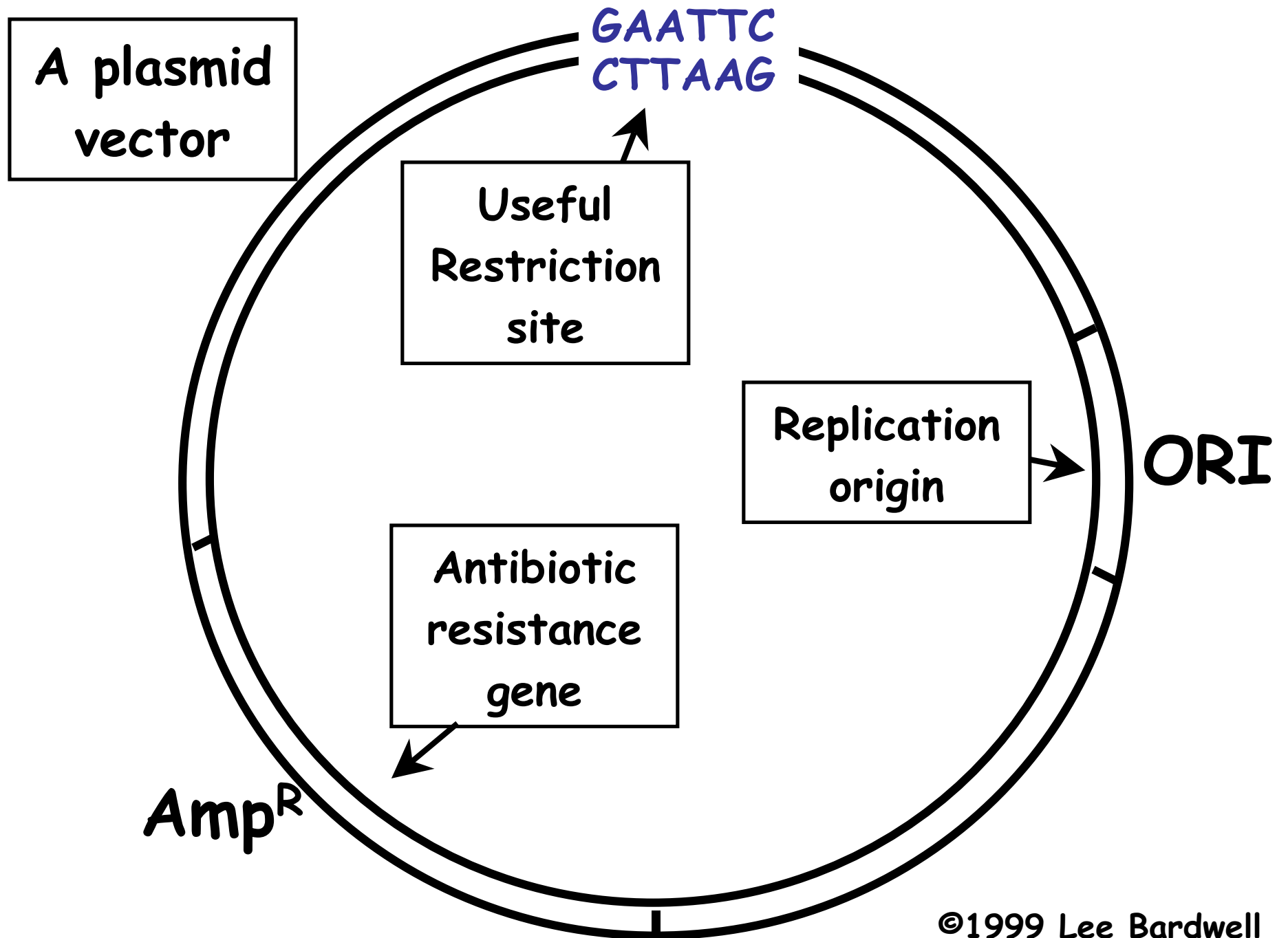
- **A carrier for recombinant DNA**
- **Typically small (<10 Kb)**
 - (1 kB = 1000 base pairs)
- **Not a chromosome but an extrachromosomal element such as a plasmid or a phage**

A useful vector...

- Can be transformed into a host cell (typically the bacterium *E. coli*)
- Contains a replication origin, so it can replicate inside that host cell
- So we can make as many copies of the cloned gene as we need

A useful vector also has...

- A selectable marker (a gene encoding resistance to an antibiotic), so that host cells containing the vector can be readily identified
- A restriction site into which foreign DNA can be inserted



What's a restriction site?

A short DNA sequence
that can be cleaved
by a restriction
enzyme

What's a restriction enzyme?

A restriction enzyme, or restriction endonuclease, cleaves DNA at a specific sequence

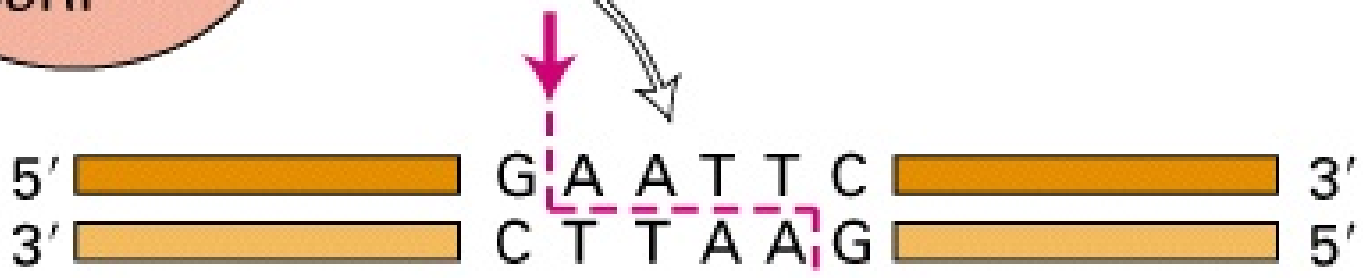
Nucleases

- Are proteins (enzymes) that cut DNA
- Exonucleases chew in from the ends
- Endonucleases cleave a within a strand
- Restriction endonucleases cut at a short, specific sequence, e.g.

5' GAATTC

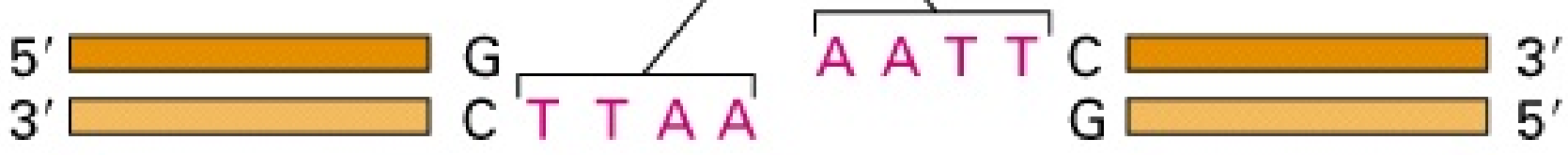
3' CTTAAG is cut by EcoRI

Restriction enzyme
EcoRI



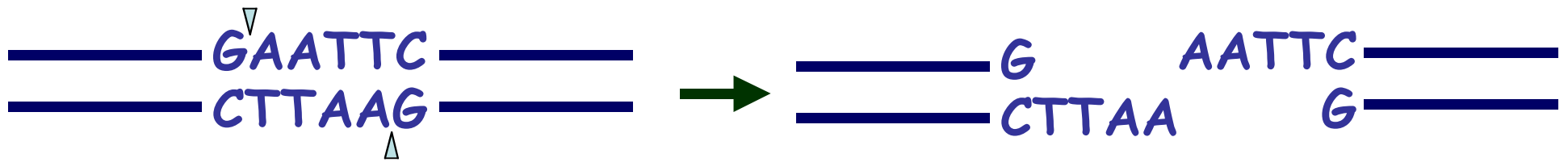
Cleavage

Sticky ends



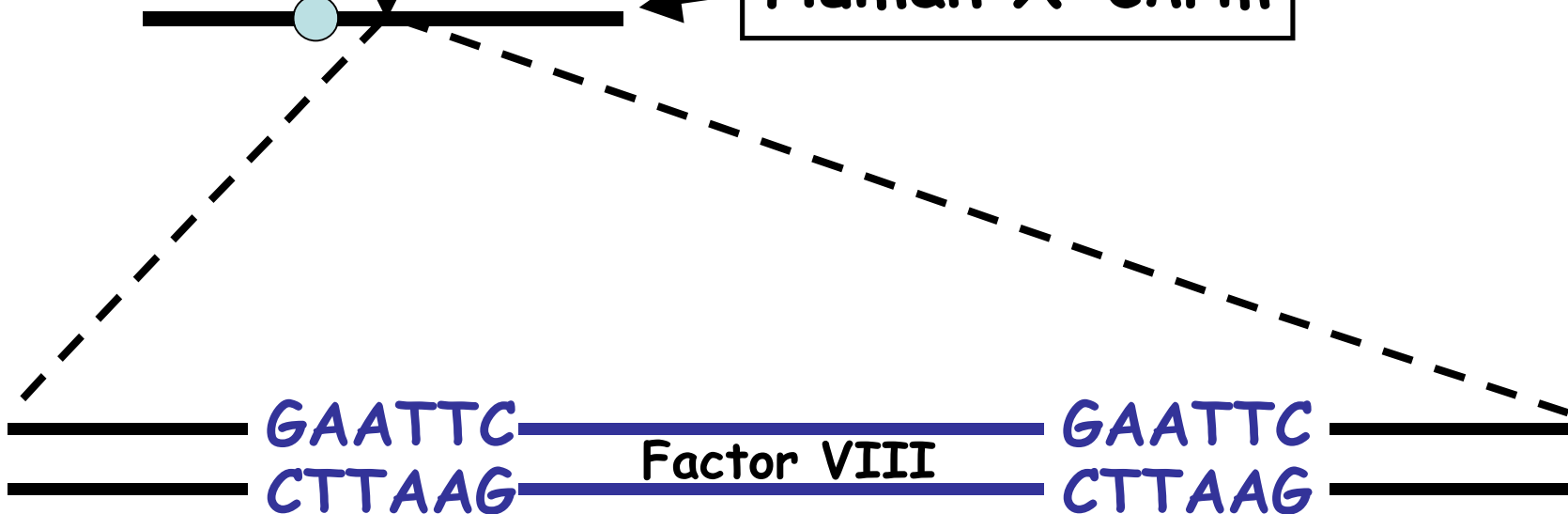
DNA Cloning

- Cleave insert and vector DNA with the same restriction enzyme to generate complementary sticky ends
- Complementary sticky ends from different DNA molecules can base pair and be covalently linked with DNA ligase --> recombinant DNA

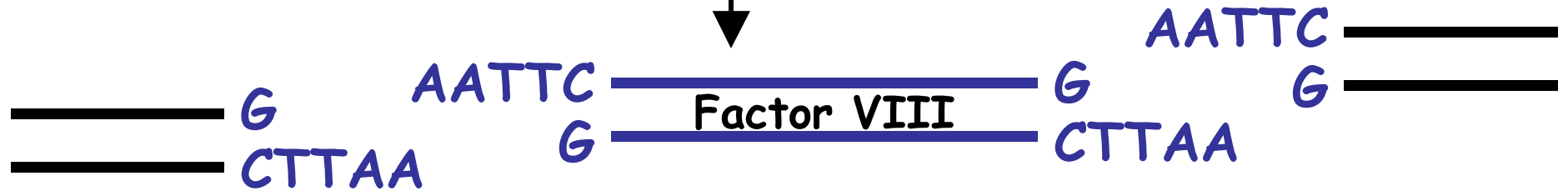


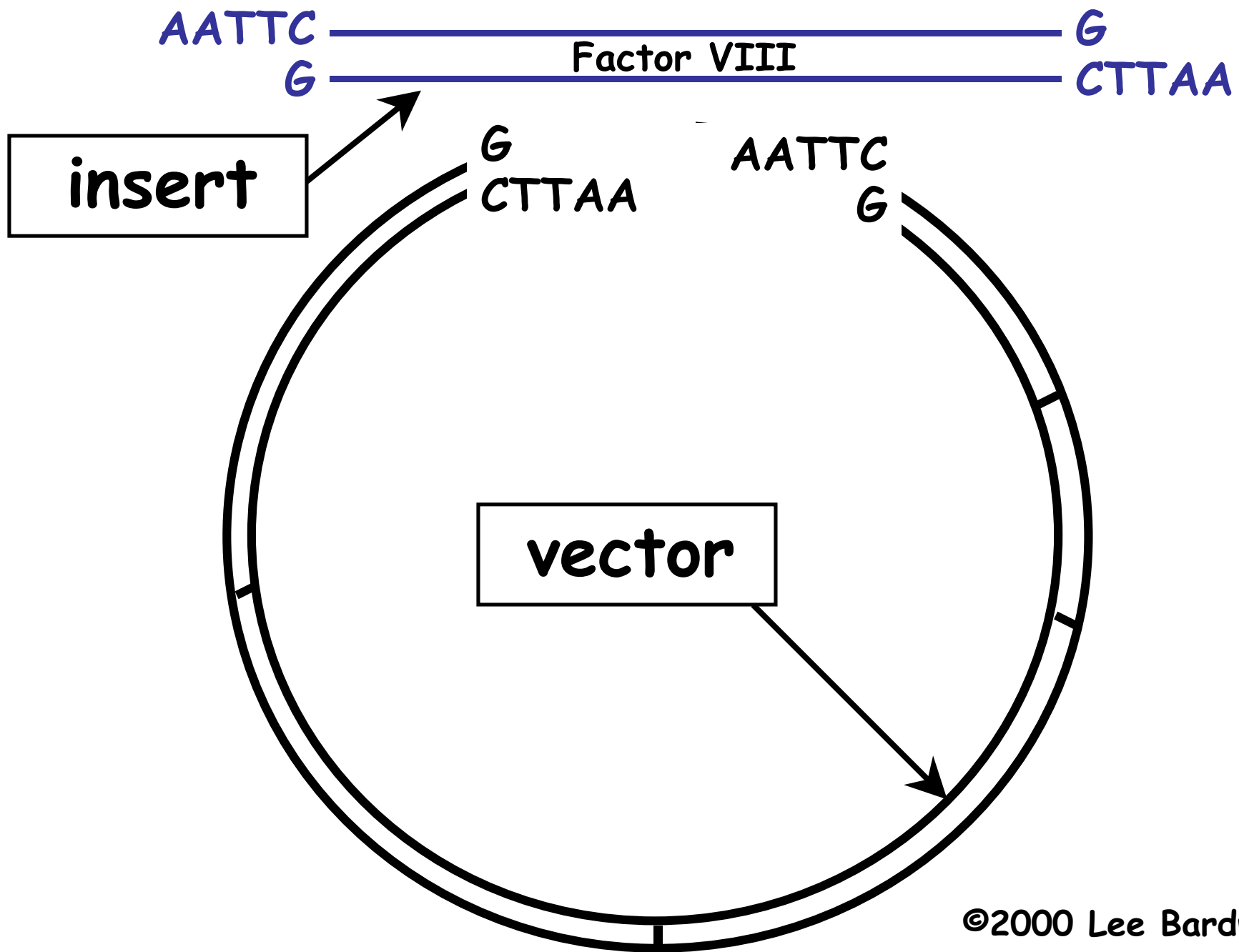
Factor VIII (Hemophilia A) locus

Human X-chrm



Cut with EcoRI restriction enzyme





Therapeutic Cloning

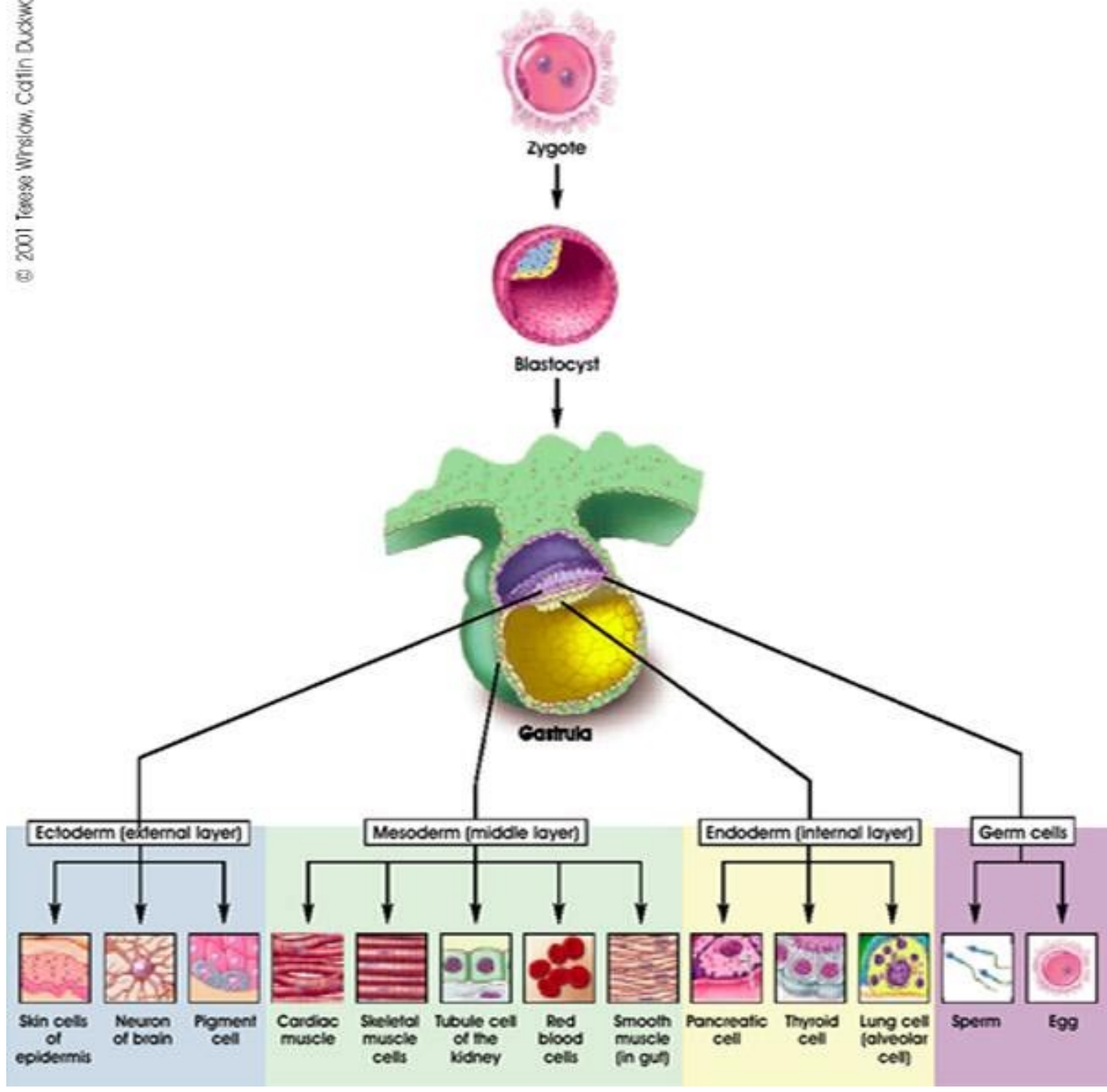
**for stem cell
research has merits**



**Ethical
Issue
of the Day #2**

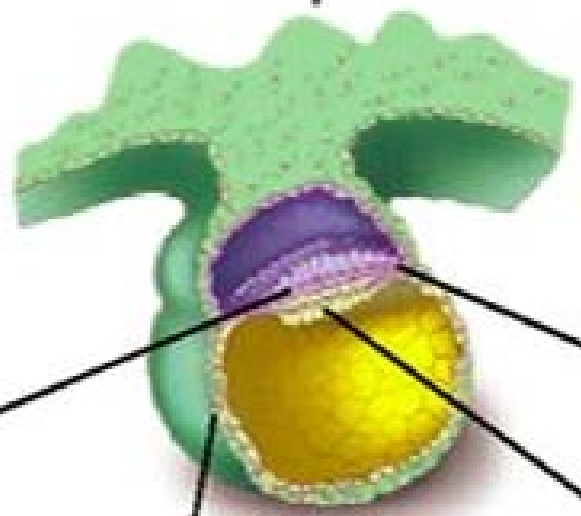
Stem Cells

- **What are stem cells?**
- **What are they good for?**
- **What is an embryo?**
- **Do all stem cells used in research come from embryos?**
- **Why are embryos useful to make stem cells?**
- **Will banning stem cell research put an end to the creation and destruction of human embryos in test tubes?**





Blastocyst



Gastrula

