Gene mapping - II: Somatic cell hybrids and FISH

Genetics 371B Lecture 11 15 Oct. 1999

Genomics

Genetic maps in humans

The trouble with humans…

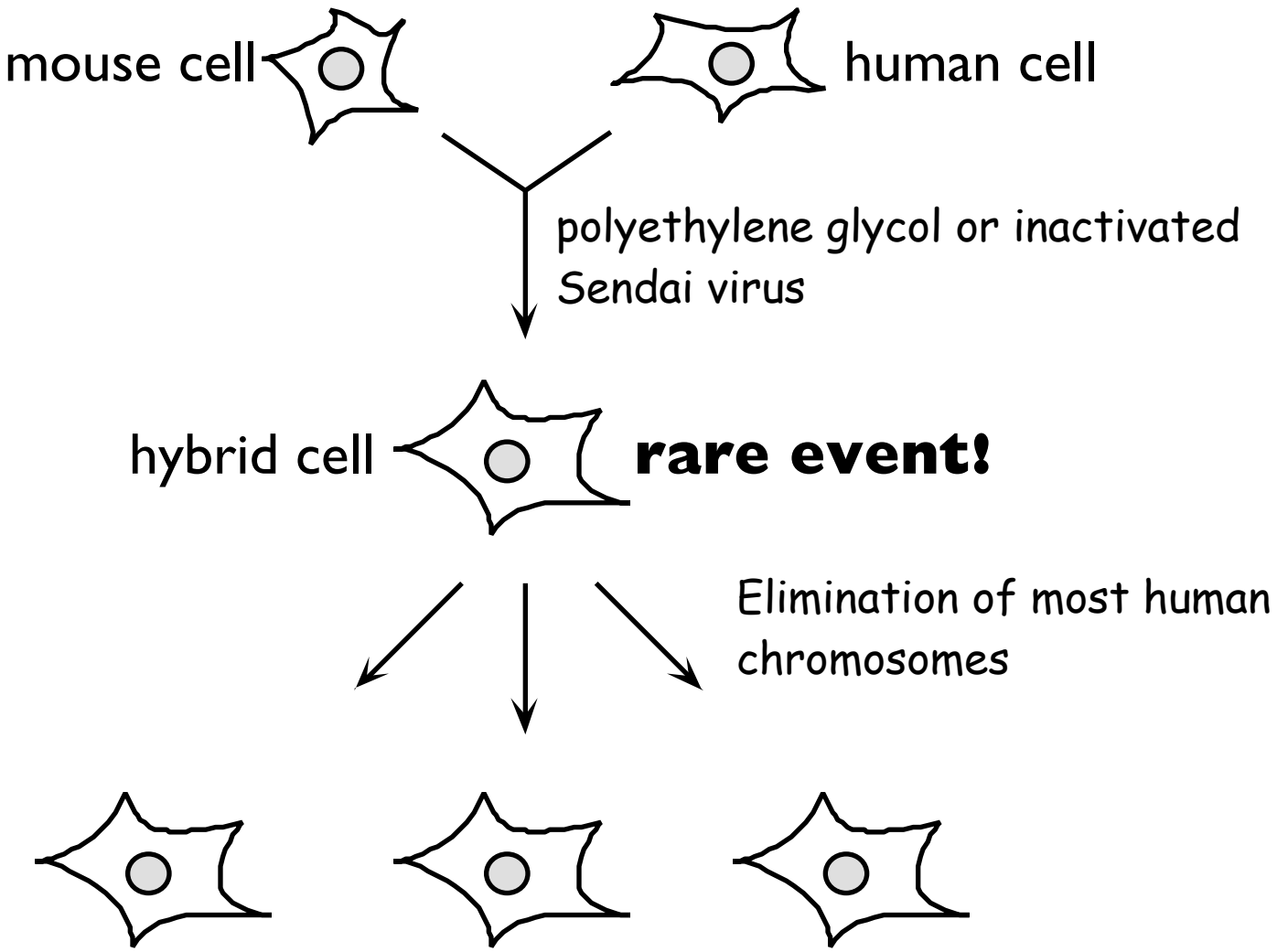
- Markers
- Crosses

Establishing linkage: which chromosome?

Recognizing chromosomes

- Size
- Staining pattern – bands

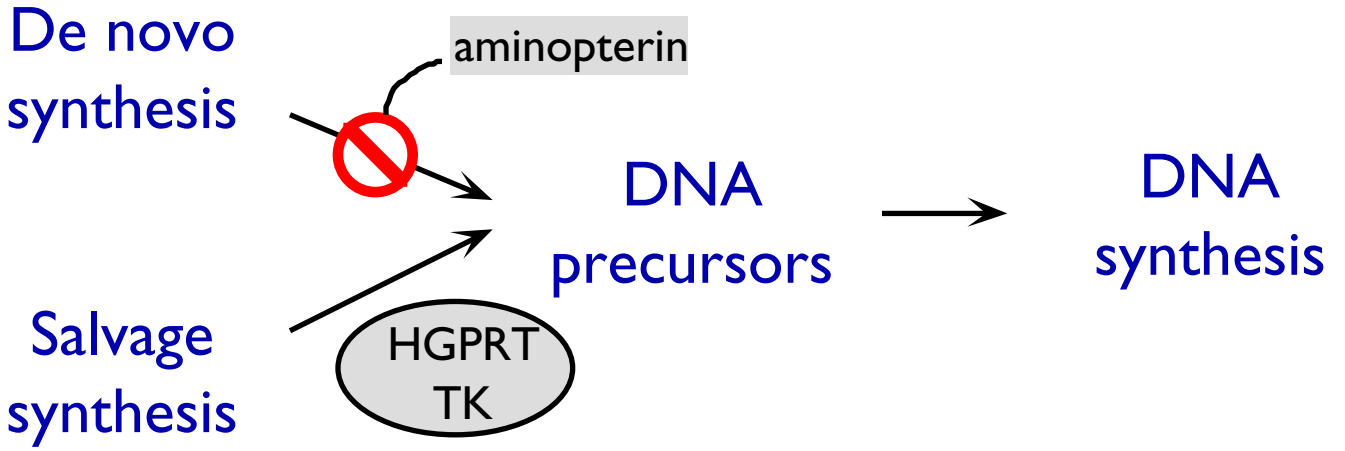
Somatic cell hybrids



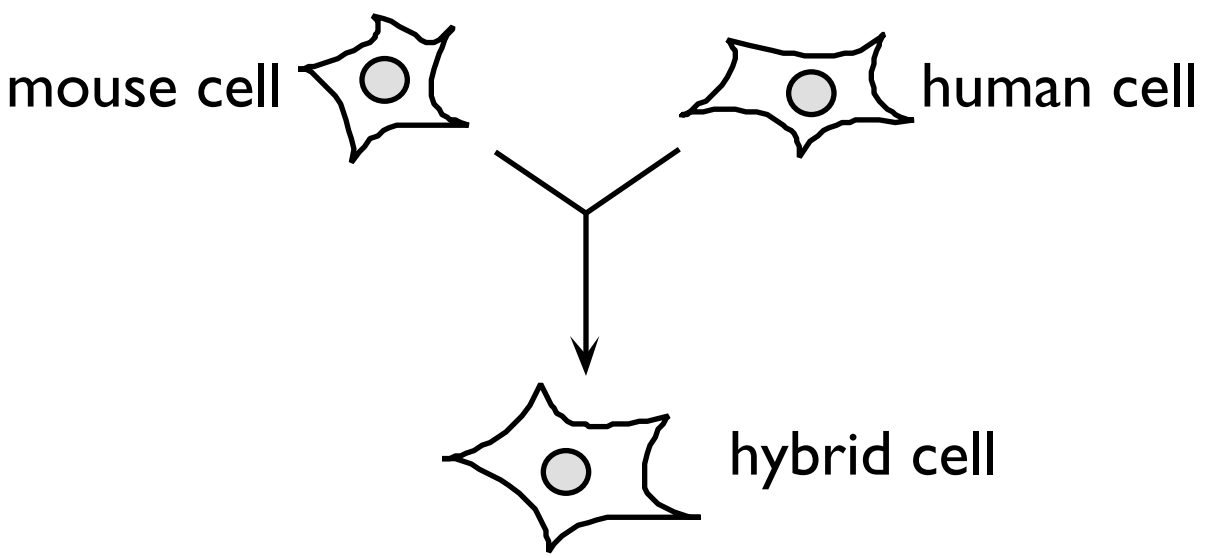
Hybrid cell lines: mostly mouse plus a few human chromosomes

How to pick out those rare fusion events?
…**selection** based on DNA precursor synthesis

Two pathways of DNA precursor synthesis:

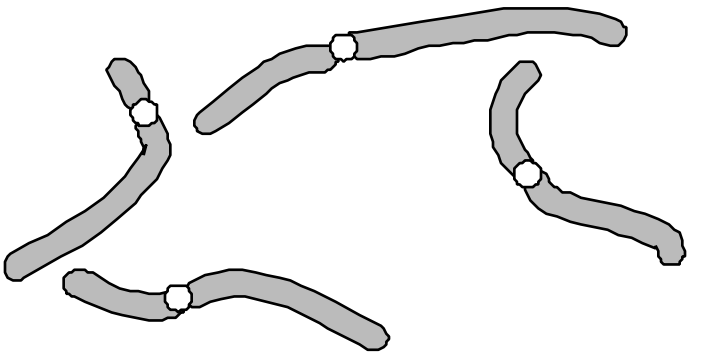


To select fusion product…



Fluorescent in situ hybridization (FISH)

Hybridize fluorescent-labeled probe to chromosome spread



…can be used in combination with somatic cell hybrids

Mapping by linkage

…linkage with respect to what?

The conventional approach – look at recombination frequency between the gene of interest and a neighboring marker gene

Conventional markers (alleles that result in overt phenotypes) are hard to come by…

But DNA sequence differences (polymorphisms) are plentiful

Therefore: construct a map of polymorphic sites

To map a gene: look at recombination frequency between the gene of interest and a neighboring polymorphic site

…so, we use DNA sequence polymorphism as just another pair of alleles – without an overt phenotype, but detectable

Useful polymorphisms

Originally – Restriction fragment length polymorphisms (RFLP)

These days – **Sequence repeat** polymorphisms

Repeated sequences constitute up to 35% of the human genome

- **Minisatellite repeats:** ~ 30 bp
- **Microsatellite repeats:** ~2–5 bp
  - ◇ Dispersed throughout the genome
  - ◇ Highly variable numbers of repeats at each location; individuals often heterozygous