Genetics 2003

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I Background from Foundations:

Introduction to Developmental Genetics

A. Model systems/ Life cycles

1) Drosophila: internal fertilization, embryo, 3 larval stages, pupa, adult. (complete metamorphosis, imaginal discs, polytene salivary gland chromosomes,)
2) *C. elegans:* internal fertilization, embryo, 4 larval stages, adult (hermaphrodite, strict cell lineage)
3) Zebrafish: external fertilization, embryo, larva, adult. (transparent embryos)

B. Meiosis

1) Spermatogenesis
2) Oogenesis

C. From genotype to phenotype: Characteristics of Alleles

1) recessive lethal genes
   How to keep a lethal line: balancers
2) Modes of inheritance
   Zygotically active genes and maternal effect genes
3) Nature of mutations
   -recessive vs. dominant
   -loss of function (lof), (amorph (null), hypomorph (weak))
   -gene dosage (haploinsufficiency (1X), hypermorph (>2 X))
   -gain of function:
   dominant active (neomorph,), dominant negative (antimorph)
   -Conditional alleles (temperature sensitive synthesis, structure, stability)

II. Identification of Gene Function by Mutagenesis

Rationale for mutant screens:

Identify genes required for a specific biological process
Isolate more mutations for a gene of interest
To obtain mutation tools for structure-function analysis
To isolate mutations in a gene identified by molecular approaches

A. Mutagens

1) Chemical
2) Transposable element
3) Irradiation
4) Targeted gene disruption
5) timing of mutagenesis. Mutagenizing spermatogonia produces clusters of sperm with identical mutation. Mutagenizing spermatids produces single

mutation. Delayed mismatch repair can lead to incomplete transmission of a mutation (mosaicism).

B. Mutagenesis Protocols

Establishing **isogenic** lines to test for recessive mutations with zygotic phenotype.

1) Drosophila
   Assay
   Balancers
   Selective markers (DTS)
2) *C. elegans* (self fertilization scheme)
3) Zebrafish (diploid scheme, shortcuts: haploid embryos can be produced with UV irradiated sperm, haploid embryos can be made diploid by: early pressure (EP) to block Meiosis II and heat shock (HS) to block the first embryonic mitosis)

C. Degree of Saturation

1) Mutation rate
2) Allele frequency
3) Poisson distribution: $f(i)= e^{-m} m^i/i!$
   (f(i) is the frequency of the i class (i allele class), m is the mean number (mean number of alleles).

The Poisson distribution describes the frequency of different types of classes that arise form sampling. This statistical tool should be used when the class sizes are small, i.e. 1, 2, 3 alleles/ gene. The Poisson distribution can not accurately describe a mutagenesis experiment because it assumes that each event (i.e. production of a mutant allele in a particular gene) occurs with equal probability. However, it is a useful tool to get an "estimate' of saturation.

4) The "one allele" class
   Variable strength of different alleles at same locus
   Mutagenicity of locus (e.g. size, mutagen)
   Overlap between maternal and zygotic gene functions
   Multiple gene functions of one gene can be separated by mutation
   Synthetic phenotypes (redundancy, more than one mutant gene involved)
   Chromosome rearrangement (translocation)

III. Genetic Characterization

A. Classification of phenotype (phenotypic classes)

B. Complementation analysis

1) Rules
   Recessive
   Reversion test for dominant gain of function mutations
   Not necessarily same phenotype
2) Complications:
   Intragenic complementation
   Second site non complementation (non-allelic non complementation

Poisoning (allele specific at both loci)
Sequestration (allele specific at at least one loci)
Combined haploinsufficiency (not allele specific)

C. Dominant Enhancer Screen

D. Suppression

1) Intragenic suppression (pseudo reversion)
   Translational
   Compensatory
   Suppression of antimorphic dominant mutations
2) Extragenic suppression
   Transcriptional
      Gene expression
      mRNA stability
   Translational
      tRNA suppressors (nonsense codon, frameshift
   Protein-protein interaction ("Dream Suppressors")
   Bypass suppression