KEYWORDS FOR BIO 2133

I. Mendelian Analysis

Mendelian analysis
It is genetic analysis based on Mendel's approach to study inheritance, they are based on his four postulates (LI slide 6).

Alleles
Alternate forms of a single gene, which determines phenotype (45). They may be dominant or recessive.

Genes
Units of inheritance represented by Mendel's unit factors (45). The fundamental physical unit of hereditary, whose existence can be confirmed by allele variants which occupies a specific chromosomal locus. A DNA sequence coding for a single polypeptide (Glossary).

Dominant allele
*An allele that is expressed in the heterozygous condition.

Recessive allele
*An allele that is not expressed in the heterozygous condition.

P
Parental generation in a monohybrid or dihybrid cross (44), in classical cases they are pure-bred with contrasting traits to be analyzed.

F1 filial
Offspring of the parental generation, will usually be all heterozygous.

F2 filial
Offspring of the F1 filial generation. This is where the phenotypical ratios are shown.

Homozygous
When both alleles are the same for one trait (45). Glossary: an individual with identical alleles for a gene or genes of interest. These individuals will produce identical gametes (with respect to the genes in question) and will therefore breed true.

Heterozygous
When both alleles are different for the one trait (45). Glossary: an individual with different alleles at one or more loci. Such individuals will produce unlike gametes and therefore will not breed true.

Hemizygous
A trait where the individual possesses one copy of a gene in an otherwise diploid cell, found when looking at X-linked inheritance where an affected male cannot be considered heterozygous (86).

Chromosome
*The location of genes and alleles in organisms, which, during cell division, separate independently (chromosome theory of inheritance). Glossary: in eukaryotes, a DNA molecule complexed with RNA and proteins to form a threadlike structure containing genetic info arranged in a linear sequence and visible during mitosis and meiosis.

Genotype
When alleles are written in pairs to represent the two unit factors present in an individual (45)

Haplotype
Glossary: a set of alleles from closely linked loci carried by an individual inherited as a unit.

Phenotype
Physical expression of a trait (45)

Monohybride (and Crosses)
A monohybrid cross is one that "mates true-breeding individuals from two parent strands, each exhibiting one of the two contrasting forms of the characterstic under study." (p. 43-44). From website: Monohybrid crosses involve a single pair of contrasting traits. The original parents are the P_1 generation (therefore purebred), and their offspring are the F_1 generation. Offspring arising from selfing (self-fertilization) the F_1 generation are the F_2 generation. The F2 generation has ratios 3:1.

Dihybrid (and Crosses)
A dihybrid cross is a two-factor cross, it analyzes two factors simultaneously (47). A dihybrid cross analyzes two strands of pure-bred parents with each two kinds of contrasting traits. P1 are pure-bred, and the F2 generation that results is 9:3:3:1. This demonstrates Mendel's postulate of independent assortment.

Trihydrid (and Crosses)
A cross analyzing three pairs of contrasting traits. Similar to dihybrid and monohybrid in how they are conducted, but in the end the F2 generation has ratios 27:9:9:9:3:3:3:1 (further demonstrating Mendel's 4^{th} postulate).

Test Cross
A way to find the genotype of an organism. Organism expressing a dominant phenotype but unknown genotype is crossed with a homozygous recessive individual. If considering one trait, a 1:1 ratio of phenotypes means the individual is heterozygous, if all dominant phenotypes then homozygous dominant (46 – 47).

Backcross
Glossary: a cross between an F1 heterozygote and one of the P1 parents (or an organism with a genotype identical to a parent).

Self Cross
Reproduction by self-fertilization.

True Breeding Strain
A strain of individuals homozygote for a certain trait (used as a P generation in a cross).

Sum Law
The probability of obtaining any single outcome, where that outcome can be achieved by two or more events, is equal to the sum of the individual probabilities of all such events (54 – 55)

Product Law
When two independent events occur simultaneously, the probability of the two outcomes occurring in combination is equal to the product of their individual probabilities of occurrence (48).

Conditional Probability
The probability of an outcome that is dependent on a specific condition related to that outcome.

Punnett Square
A diagram used to visualize the genotype and phenotype results from cross breeding (46)

Chi-square analysis
x = sum of (observation – expected)^2/ expected

X Chromosome
One of the sex chromosomes, larger than the Y chromosome.

Y Chromosome

One of the sex chromosomes, where XY pairing designates male. It lacks copies of most genes present on the X chromosome.

X-linkage
Inheritance where the gene is present on the X chromosome

Y-linkage
Inheritance where the genes is present on the Y chromosome

Meiosis
Glossary: the process of cell division in a gametogenesis or sporogenesis during which diploid number of chromosomes in reduced to the haploid number.

Locus
The location on a given chromosome where any particular gene occurs (54)

Pedigree
A family tree indicating the presence or absence of the trait in question for each member of each generation

II. Mendelian Analysis Reloaded

Allele
A variant of a gene.

Wild Type Allele
* The phenotype of a certain trait that is the one most primarily seen in nature or in the wild. It is usually (but not always) dominant.

Incomplete Dominance
Expressing a heterozygous phenotype that is distinct from the phenotype of either homozygous parent (also called partial dominance). Ex: red and white flowers make pink flowers.

Codominance
Condition in which the phenotypic effects of a gene's alleles are fully and simultaneously expressed in the heterozygote. Ex: blood type

Multiple Alleles
In a population of organisms, three or more alleles of the same gene are present. Ex: blood type.

Dominant Lethal Allele
* A dominant allele that causes death in the organism – in the homozygous state may cause death in utero. Ex: Hungington's Disease.

Recessive Lethal Allele
* A recessive allele that causes death in the organism – usually the phenotype is only expressed as lethal when homozygous. Ex: Tay-Sachs Disease.

Complementary Gene Interaction
*Gene interaction where one gene manufactures a molecule, which is then modified by another gene. There is a biochemical pathway involved. To express the phenotype, two particular allees (usually dominant) must be present. Usually causes a 9:7 cross. Ex: Only flowers with A-B- turn out purple, all others are white.

Epistasis
One gene influences or interfere with the expression of another gene leading to a specific phenotype. Ex: a gene modifies the black pigment to agouti. Examples of ratios, 9:3:4 (recessive) and 12:1:3 (dominant).
Nonreciprocal interaction between nonallelic genes such that one gene influences or interferes with the expression of another gene, leading to a specific phenotype.

Gene Interaction
Any mechanism where genes interact to modify the 9:3:3:1. Production of novel phenotypes by the interaction of alleles of different genes.

Pleiotropy
Condition in which a single mutation causes multiple phenotypic effects.

X-linkage
The pattern of inheritance resulting from genes located on the X chromosome. Examples are haemophilia, colour blindness.

Sex-Influenced Inheritance
Phenotypic expression that is conditioned by the sex of the individual. A heterozygote may express one phenotype in one sex and the alternate phenotype in the other sex. Can be due to hormonal profile. Example: pattern baldness in humans.

Conditional (Temperature-Sensitive) Mutation
A mutation that is expressed only under a certain condition, that is, a wild-type phenotype is expressed under certain (permissive) conditions and a mutant phenotype under other (restrictive) conditions. Ex: dark spots in Siamese cats.

Expressivity
The degree of range in which a phenotype for a given trait is expressed.

Genetic Suppression
A gene suppressed the phenotypic expression of another gene (ratio 13:3).

Penetrance
The frequency, expressed as a percentage, with which individuals of a given genotype manifest at least some degree of a specific mutant phenotype associated with a trait.

Phenotypic expression
The expression of a genotype (thus the expression of a phenotype).

Human Genome Project

Human genome project
Research that had goals to: 1) set out to create a map of the human and other species' genomes, 2) find location of all genes, 3) compile lists of expressed genes and nonexpressed genes, 4) discover function of all genes, 5) ID proteins encoded by genes and their functions, 6) compare genes and proteins between species, 7) analyze DNA differences between genomes, 8) setup and manage databases on genomes discovered.

Genomes
The set of hereditary information encoded in the DNA of an organism, including both the protein-coding and the non-protein-coding sequences.

DNA
A macromolecule usually consisting of polymers of nucleotides comprising antiparallel chains in which the sugar residues are deoxyribose and which are held together by hydrogen bonds. The primary carrier of genetic info

Genomics
A subdiscipline of the field of genetics generated by the union of classical and molecular biology with the goal of sequencing and understanding genes, gene interaction, genetic elements and the structure of genomes.

Proteomics
The study of the expressed proteins present in a cell at a given time.

Complete Linkage
A condition in which two genes are located so close to each other that no recombination occurs between them

Crossing over
The exchange of chromosomal material (parts of chromosomal arms) between homologous chromosomes by breaking and reunion. The exchange of material between nonsister chromatids during meiosis is the basis of genetic recombination.

Linkage
The condition in which genes have their loci present on the same chromosome, causing them to be inherited as a unit, provided that they are not separated by crossing over during meiosis.

Linkage group
Genes that are located on the same chromosome and show "linkage" to one another.

Linkage ratio
* The ratio between frequency of crossing over and the amount of offspring or results all together.

Parental (noncrossover) gamete
A gamete whose chromosomes have undergone no genetic recombination, that look like the parental gametes.

Recombination
The process that leads to the formation of new gene combinations on chromosomes.

Recombination (crossover) gamete
A gamete containing a new combination of genes produced by crossing over during meiosis

Centimorgan (cM)
A unit of distance between genes on chromosomes representing 1 percent crossover between two genes. Equivalent to one map unit (mu).

Chiasma (plural chiasmata)
The crossed strands of nonsister chromatids seen in diplotene of the first meiotic division. Regarded as the cytological evidence for exchange of chromosomal material, or crossing over.

Map Unit (mu)
See centimorgan

Single Crossover (SCO)
* Only one event of crossing over.

Double Crossover (DCO)
Two separate events of chromosome breakage and exchange occurring within the same tetrad

Coefficient of Coincidence (C)
A ratio of the observed number of double crossovers divided by the expected number of such crossovers.

Interference
A measure of the degree to which one crossover affects the incidence of another crossover in an adjacent region of the same chromatid. Negative interference increases the chance of another crossover; positive interference reduces the probability of a second crossover event.

Tetrad
The four chromatids that make up paired homologs in the prophase of the first meiotic division. In eukaryotes with a predominant haploid stage (some algae and fungi), tetrad denotes the four haploid cells produced by a single meiotic division.

Two point mapping
*Mapping genes on a chromosome looked solely at two genes, thus double crossovers are ignored

Three point mapping
*Mapping genes on a chromosome looked solely at three genes, therefore double crossovers are taken into consideration

Biotechnology

Biotechnology
- Glossary: commercial and/or industrial processes that utilize biological organisms or products
- Notes: analyze DNA sequences, measure gene expression, construct molecules, produce transgenic organisms, produce medicines/drugs/beer/milk, bioremediation, biological weapons, genetic engineering, gene therapy, genetic testing.
- Info: Whereas ***modern biotechnology*** manipulates the genes of organisms and inserts them into other organisms to acquire desired traits (i.e. production of human therapeutic drugs, infection resistant crops, transgenic fish, gene cloning, etc..)., ***traditional biotechnology*** uses the processes of organisms for desired outcomes (i.e. fermentation, bioremediation, isolation of specific products, use of specific enzymes etc...).
  In its purest form, the term biotechnology refers to the use of living organisms or their products for specific objectives, such as for the production of food, drink, medicine, or for other benefits to the human race, or other animal species. It can also be used to solve problems and conduct research.

Recombinant DNA
- Glossary: A term also applied to the technology associated with the use of DNA molecules produced by in vitro ligation of DNA from two different organisms.
- Slides/*: - To create recombinant DNA, one has to use restriction enzymes to cut up the DNA and recombine it. Example of steps: 1) Cleavage with EcoRI, 2) Annealing allows recombinant DNA molecules to form complementary base pairing. The two strands are not covalently bonded, 3) DNA ligase seals the gap

Restriction enzyme
- A bacterial nuclease that recognizes specific nucleotide sequences in a DNA molecule, often a PALINDROME (ex race car) as seen in both directions of the double strands, and cleaves or nicks the DNA at those sites. Provides defence for invading DNA segments in bacteria and is useful in the construction of recombinant DNA molecules.
- Slides/*: - enzymes often cleave in an offset manner, producing "sticky ends". These sticky ends allow for strands to H-bond without being covalently bonded (DNA Ligase covalently bonds after).
  - Examples: EcoRI (GAATTC), HindIII (AAGCTT), BamHI (GGATCC)

Restriction site
*The site on DNA (a sequence, usually palindromic) where a restriction enzyme will cleave.

Restriction fragment
*Fragment of DNA generated by restriction enzymes.

Cloning vector
- Glossary: In recombinant DNA, an agent such as a phage or plasmid into which a foreign DNA segment will be inserted and utilized to transform host cells.
- Slides: Vectors are carrier DNA molecules that can replicate DNA fragments in a host cell.
  - Vectors must be able to replicate independently and should have several restriction enzyme sites to allow insertion of a DNA fragment.
  - Ex: plasmids, phage vectors, cosmid vectors, bacterial artificial chromosomes, expression vectors.

Bacterial artificial chromosome (BACs)
- Slides: Based on F factor and can carry up to 300 kb of inserted DNA.
- Wikipedia: DNA construct, based on a functional fertility plasmid (or F-plasmid), used for transforming and cloning in bacteria, usually *E. coli*.[1][2] F-plasmids play a crucial role because they contain partition genes that promote the even distribution of plasmids after bacterial cell division.

Cosmid
- Glossary: A vector designed to allow cloning of large segments of foreign DNA. Cosmids are composed of the *cos* sites of phage lambda inserted into a plasmid. In cloning, the recombinant DNA molecules are packaged into phage protein coats and after infection of bacterial cells, the recombinant molecule replicates and can be maintained as a plasmid.

- Slides: vectors are created by combining parts of lambda phage and parts of plasmids.
  - contain the cos sites of lambda, necessary for packaging of phage DNA into phage particles
  - once inside the bacterial cell, cosmids replicate as plasmids
  - cosmids can carry almost 50 kb of inserted DNA.

Phage vector

- Slide: the central third of lambda phage vectors can be replaced with foreign DNA without affecting the ability to infect cells and replicate. They can carry up to 20 kb of cloned DNA.
  - Lambda phage has DNA (central gene cluster). Central region is removed by restriction enzyme digestion
  - Lambda arms are produced. Insertion and ligation of foreign DNA occurs between arms.
  - After in vitro packaging, recombinant vital particle carrying foreign insert is capable of infecting host cell and replicating to form plaques.

Plasmid

- Glossary: An extrachromosomal circular DNA molecule that replicates independently of a single bacteriophage.
  *It can be used for biotechnology, where the plasmid contains recombinant DNA and genes of interest
- Slides/*: Plasmids used for DNA cloning usually have been engineered to contain:
  - a number of convenient restriction sites (polylinker)
  - a marker gene to select for its presence in the host cell (Ex: ampicillin resistance, lacZ gene)
  - E.coli with plasmid without insert can metabolize Xgal and will form blue colony (due to lacZ)
  Polylinker is cut with restriction enzyme, cutting this gene.
  Recombinant plasmid cannot metabolize gal and will form white colony.

Polylinker

Glossary: a segment of DNA that has been engineered to contain multiple sites for restriction enzyme digestion.
Polylinkers are usually found in engineered vectors such as plasmids.

Agarose gel

*A gel made out of agarose, a porous gel where DNA or proteins can migrate towards an opposite charged electrode
Used in gel electrophoresis.

Gel electrophoresis separates charged molecules in an electric field.
DNA is a charged particle
Small fragments migrate faster than bigger fragments.

Polymerase chain reaction

Glossary: (PCR) a method for amplifying DNA segments that depends on repeated cycles of denaturation, primers, and DNA polymerase-directed DNA synthesis.

Slides: - makes DNA copies without host cells
- copies a specific DNA sequence through in vitro reactions that can amplify target DNA sequences present in very small quantities
- requires two oligonucleotide primers, one complementary to the 3' end of one strand of the DNA to be amplified and one complementary to the 3' end of the other strand.
- You need: DNA (genomic or cDNA), two primers (sense and antisense), excess of nucleotidues, Taq polymerase, buffer with magnesium chloride, thermocycler
- Three steps:
1) Denaturation: at 94 degrees Celsius, H-bonds break
2) Primer annealing: at 37degrees to 65 degrees
3) Extension: at 72 degrees Celsius, primers are extended using Taq polymerase
- steps are repeated over and over again using a thermocycler to amplify DNA exponentially

Primers

Glossary: In nucleic acids, a short length of RNA or single-stranded DNA required for the initiating synthesis directed by polymerases.

*for PCR, have to be oligonucleotides, and usually we have a sense of the sequence beforehand (or use random hexomer primers)

Polymerase

Glossary: Enzymes that catalyze the formation of DNA and RNA from deoxyribonucleotides and ribonucleotides.

*During PCR, Taq polymerase is used. It is isolated from Thermus Aquaticus bacteria, who live in Geysers at average temperature of 86 degrees Celsius. They can withstand the temperatures of the thermocycler. They are stable at 95 degrees C for one hour.

cDNA library

Glossary: a collection of cloned cDNA sequences. (cDNA: DNA synthesized from an RNA template by the enzyme reverse transcriptase).

Slides: - isolating mRNA from cells
- Synthesizing the complementary DNA using reverse transcriptase
- cloning the DNA molecules into a vector

Genomic library

Glossary: a collection of clones that contains all the DNA sequences of an organism's genome.

Slides: Ideally contains one copy of all sequences in the genome of interest

Genomic libraries are constructed by cutting genomic DNA with a restriction enzyme and ligating the fragments into vectors

The choice of vector usually depends on the size of the genome

Probe

Glossary: A macromolecule such as DNA or RNA that has been labelled and can be detected by an assay such as autoradiography or fluorescence microscopy. Probes are used to identify target molecules, genes, or gene products.

Slides: - complementary to part of a gene are used to screen a library to recover clones of a specific gene
- the colony corresponding to the one the probe identified on the filter is identified and recovered
- Steps: 1) Colonies of the library are overlaid with a DNA-binding filter
2) Colonies are transferred to filter, then lysed, and DNA is denatured
3) Filter is placed in a heat-sealed bag with a solution containing the labelled probe; the probe Hybridizes with denatured DNA from colonies
4) filter is rinsed to remove excess probe, then dried; X-Ray film is placed over the filter for autoradiography
5) Using the original plate, cells are picked from the colony that hybridized to the probe
6) Cells are transferred to a medium for growth and further analysis

Southern blot

Glossary: developed by Edwin Southern, a technique in which DNA fragments produced by restriction enzyme digestion are separated by electrophoresis and transferred by capillary action to a nylon or nitrocellulose membrane. Specific DNA fragments can be identified by hybridization to a complementary radioactively labelled nucleic acid probe using the technique of autoradiography.

Slides: - can be used to identify which clones in a library contain a given DNA sequence, characterize the size from restriction digest, determine whether a clone contains all or parts of a gene, ascertain size and sequence organization of gene of interest
- Steps:
1) DNA samples cut with restriction enzymes are loaded on agarose gel for electrophoresis
2) DNA is separated by electrophoresis
3) DNA-binding filter, paper towels and weight are placed on gel; buffer passes upward through sponge by capillary action, transferring DNA fragments to filter
4) After DNA is denatured, filter is placed in heat-sealed bag with solution with probe; hybridization
5) Filter is washed to remove unbound probe, then dried; Xray film is applied to autoradiography

Northern blot

Glossary: an analytical technique in which RNA molecules are separated by electrophoresis and transferred by capillary action to a nylon or nitrocellulose membrane. Specific RNA molecules can then be identified by hybridization to a labelled nucleic acid probe.

Chain termination DNA sequencing
*Sequencing DNA where dideoxyribonucleotides are used so that segments are created where the last base is known.
- Slides: - the most common method of DNA sequencing s dideoxy chain termination sequencing, developed by Sanger
  - Template = any DNA fragment (from PCR, plasmid with insert, etc.)
  - Perform sequencing reactions with ddATP, ddTTP, ddGTP, ddCTP
  - electrophorese each reactions in a different lane of gel

dNTP
Deoxyribonucleotide triphosphates

ddNTP
Dideoxyribonucleotide triphosphate

Automated DNA sequencing
*using fluorescent probes for DNA sequencing

Bioinformatics
Glossary: the design and application software and computational methods for the storage, analysis, and management of biological information such as nucleotide or amino acid sequences.