Methods and Logic: Gregor Mendel Experiments in Plant Hybridization Cloe Pogoda 2-19-14

Mendel



the formulation and development of hybrids has yet been successfully formulated can hardly astonish anyone who is acquainted with the extent of the task and who can appreciate the difficulties with which experiments of

Mendel

- Johann Mendel was born into an ethnic German family in 1822.
- He spent his early youth in that rural setting, until age 11, when a local schoolmaster who was impressed with his aptitude for learning recommended that he be sent to secondary school to continue his education.
- Upon recommendation Mendel entered the Augustinian St Thomas's Abbey and began his training as a priest in 1843. He took the name Gregor upon entering religious life.
- In 1854 Mendel was authorized to conduct his investigation in the monastery's 2 hectares (4.9 acres) experimental gardens.
- In 1865, Mendel delivered two lectures on his findings to the Natural Science Society in Brno, who published the results of his studies in their journal the following year, under the title *Experiments on Plant Hybrids*
- Mendel died in 1884, at the age of 61 from chronic nephritis.

The world in the 1860s

- The American Civil War lasted from 1861-1865.
- Speed of light was measured by Foucault in 1862.
- Dmitri Mendeleev develops the modern periodic table in 1869.
- Alfred Nobel creates dynamite in Sweden,
 - His fortune was used posthumously to institute the Nobel Prizes.
- Florence Nightingale founds school for nurses in 1860.
- Maxwell publishes his equations that quantify the relationship between electricity and magnetism in 1862.



Background

- Agriculturists had long known that the dominant phenotype was often inherited by the progeny.
- Mendel wished to discover the laws that governed inheritance.
 - "It requires indeed some courage to undertake a labor of such far-reaching extent; this appears. However, to be the only right way by which we can finally reach the solution of a question the importance of which cannot be overestimated in connection with the history of the evolution of organic forms."

Questions the Paper Addresses

- Darwin and Wallace had previously described their observations on how varieties arose from species, but a deeper understanding was not yet available until Mendel was able to elucidate the laws of inheritance.
- Mendel undertook 8 years of experimentation to understand what is now the fundamentals of inheritance.



- Mendel first had to select the appropriate plants for his experimentation:
 - *The plants must possess constant differentiating characteristics.*
 - The hybrids of such plants must, during the flowering period, be protected from the influence of all foreign pollen, or be easily capable of such protection.

Experimental Model

- Mendel studied the garden pea (*Pisum sativum*) for several reasons.
 - Peas were available from seed merchants in a wide array of distinct shapes and colors that could be easily identified and analyzed.
 - Peas can either self-pollinate or be cross-pollinated.
 - Peas are inexpensive and easy to obtain, take up little space, have a short generation time, and produce many offspring.



Experimental Model

• Pea plant anatomy:



- Mendel obtained lines of plants, which he grew for two years to make sure that they were pure.
- A pure line is a population that breeds true for the particular character being studied.
- Mendel had effectively established a control experiment by producing a fixed baseline for his future studies so that any changes observed subsequent to deliberate manipulation in his research would be scientifically meaningful.



| | Characteristic #3 | Characteristic #6 | Characteristic #2 | Characteristic #1 | Characteristic #4 | Characteristic #5 | Characteristic #7 |
|----|-------------------|------------------------|----------------------|------------------------|------------------------------|----------------------|-------------------|
| | Flower color | Flower position | Seed color | Seed shape | Pod shape | Pod color | Stem length |
| Р | Purple | Axial × Terminal | Yellow × Green | Round × Wrinkled | Inflated × Constricted | Green × Yellow | Dwarf |
| F, | Purple | Axial | Yellow | Round | Inflated | Green | Tall |

- Mendel chose to pick two differentiating characteristics and unite them by cross fertilization.
- He utilized male and female plants to cross and make his first generation.
- Some possible experimental errors that he mentioned were:
 - Bug contamination: *Buchus pisi*
 - Unwanted pollination from other sources
 - Pot planted vs garden grown plants



- Mendel knew from previous experiments that hybrids, as a rule, are not exactly intermediate between the parental species.
- Mendel begins to use the terms dominant and recessive to describe traits that are either seen in the F1 hybrids or not.
- Mendel also describes that he sees no way to determine by looking at a hybrid if the dominant phenotype came from the maternal or paternal plant.



- Once Mendel had produced the hybrids, he proceeded to perform self fertilization and count the number of plants or seeds that had the defining characteristics of the parental strains.
- Parental strands AA & aa
- Hybrids (F1): Aa & Aa
- F2 generation: AA & Aa & aa, which yields a 3:1 ratio
- Mendel performed many experiments and consistently obtained ratios very close to 3:1 for *dominant* to *recessive* phenotypes.





| | Parental phenotype | F ₁ | F ₂ | F ₂ ratio |
|----|-------------------------------------|----------------|---------------------------|----------------------|
| 1. | Round×wrinkled seeds | All round | 5474 round; 1850 wrinkled | 2.96:1 |
| 2. | $Yellow \! \times \! green \ seeds$ | All yellow | 6022 yellow; 2001 green | 3.01:1 |
| 3. | Purple×white petals | All purple | 705 purple; 224 white | 3.15:1 |
| 4. | $Inflated \times pinched \ pods$ | All inflated | 882 inflated; 299 pinched | 2.95:1 |
| 5. | $Green \times yellow \ pods$ | All green | 428 green; 152 yellow | 2.82:1 |
| 6. | Axial×terminal flowers | All axial | 651 axial; 207 terminal | 3.14:1 |
| 7. | Long 	imes short stems | All long | 787 long; 277 short | 2.84:1 |

- Mendel took great care in executing his experiments.
 - He ensured that the pods remained on the plants until they were thoroughly ripened and fully dried.
 - He also made sure that short plants were moved so that they did not become dwarfed by their taller brethren and die from a lack of sun.



 The study of the individual "selfings" revealed that underlying the 3:1 phenotypic ratio in the F2 generation was a more fundamental 1:2:1 genotypic ratio.



- The ratio of 3:1 that Mendel obtained is consistent with but better described by 1:2:1, which is revealed by using a Punnett square.
- Mendel continues to breed his plants through subsequent generations and sees that the proportions continued to hold true.



- He then proceeded to perform crosses where each plant had more than one differing characteristic.
- He concludes:
 - "that the offspring of the hybrids in which several essentially different characters are combined exhibit the terms of a series of combinations, in which the developmental series for each pair of differentiating characters are united. The relation of each pair of different characters in hybrid union is independent of the other differences in the two original parental stocks"

| | BbSs x BbSs | | | | | | | |
|----|-------------|------|------|-------|--|--|--|--|
| | BS | Bs | bS | bs | | | | |
| BS | BBSS | BBSs | BbSS | BbSs | | | | |
| Bs | BBSs | BBss | BbSs | Bbss | | | | |
| bS | BbSS | BbSs | bbSS | bbSs | | | | |
| bs | BbSs | Bbss | bbSs | bb ss | | | | |



$$\frac{3}{4} \times \frac{3}{4} = \frac{9}{16} \text{ round yellow}$$
$$\frac{3}{4} \times \frac{1}{4} = \frac{3}{16} \text{ round green}$$
$$\frac{1}{4} \times \frac{3}{4} = \frac{3}{16} \text{ wrinkled yellow}$$
$$\frac{1}{4} \times \frac{1}{4} = \frac{1}{16} \text{ wrinkled green}$$

Conclusions from the Results

- Mendel was able to correctly conclude the ratios of inheritance.
- He developed the terms *dominant* and *recessive*.
- He also begins to understand the functional nature of germ cells and separate alleles.
 - "The peas hybrids form egg and pollen cells which, in their constitution represent in equal numbers all constant forms which results from the combination of the characters united in fertilization."





Conclusions

- The existence of genes.
- Genes are in pairs. Mendel's reasoning here was obvious and due to his results from the F1 population with dominant and recessive phenotypes.
- The principle of segregation.
- Gamete content.
- Random fertilization.



Mendel's Laws

- The two members of a gene pair segregate from each other in meiosis; each gamete has an equal probability of obtaining either member of the gene pair.
- The law of independent assortment; unlinked or distantly linked segregating gene pairs assort independently at meiosis.



Concluding thoughts

- Mendel described basic genetic inheritance, and pioneered the terms *recessive* and *dominant*.
- He leaves the understanding of the coloring of ornamental flowers to future experiments.
- He also notes that there are great differences between plants and animals that are bred in captivity to animals that are in the wild.



Difficulties with Mendel's work

- Mendel did not promote his own work, so his achievements were not recognized in his lifetime.
- Very hard to reproduce. Subsequent crosses had difficulty with low fertility and phenotypes that didn't fall into a neat dominant/recessive patterns.
- He was unable to understand the genetic inheritance processes of quantitative, dilution, sex-linked and crossover events.



Rediscovery of Mendel's work

- Mendel and his laws were "rediscovered" by Hugo Marie de Vries, Karl Franz Joseph Correns, and Erich Tschermak von Seysenegg.
- Between the 1860s and 1900 developments in the study of cell division, fertilization, and the behavior of subcellular structures had established a new framework capable of accommodating Mendel's "ratios and numbers."
- William Bateson in the 1890s coined and popularized many of the terms now used by geneticists, including the words "genetics", "allelomorph" (allele), "zygote," "homozygote," and "heterozygote."
- In 1902 Bateson published *Mendel's Principles* of Heredity.



Controversy

- In 1902, two years after Mendel's work was rediscovered, W. F. R. Weldon suspected that Mendel's results were very close to expected values and tested this suspicion with Pearson's newly developed χ² test.
- He concluded that Mendel's observed ratios were astonishingly close to his expectations.



Controversy

 Ronald Fisher in 1936 examined Mendel's work using statistics and claimed that Mendel must have "cooked" his results because his ratios were too good.



How much did Darwin influence Mendel?

- Darwin did not have an explanation for how traits could be preserved over the succeeding generations. At the time, the prevailing theory of inheritance was that the traits of the parents were *blended* in the offspring.
- Mendel had the answer to Darwin's problem. Traits were not blended, but inherited whole.
- Mendel's genetics was combined with Darwin's original theory to give us modern Neo-Darwinism.
- Mendel mentions Darwin four times in his writing, so the overall influence is unclear.



Discussion

- It is fairly impressive that Mendel was able to correctly elucidate the general laws of inheritance.
- Whether or not Mendel did fudge his results, he did correctly predict the ratio of genetic inheritance.
- As modern scientists we still utilize this basic understanding and have added to it throughout the years.
- Along with Darwin and Wallace, Mendel contributed hugely to shifting the paradigm surrounding science and to promoting the current scientific understanding and methodology employed today.

