Mendel's Experiments and Punnett Squares

Inquire: Mendel’s Experiments and the Laws of Probability

Overview

Genetics is the science of heredity. Austrian monk Gregor Mendel set the framework for genetics long before chromosomes or genes had been identified, even at a time when meiosis was not well understood. Working with garden peas, Mendel found that crosses between true-breeding parents (P) that differed in one trait produced first generation (F1) offspring that all expressed the trait of one parent. Mendel used the term dominant to refer to the trait that was observed, and recessive to denote that non-expressed trait, or the trait that had “disappeared” in this first generation. When the F1 offspring were crossed with each other, the F2 offspring exhibited both traits in a 3:1 ratio. Other crosses generated the same 3:1 ratio in the F2 offspring. By mathematically examining sample sizes, Mendel showed that genetic crosses behaved according to the laws of probability, and that the traits were inherited as independent events. In other words, Mendel used statistical methods to build his model of inheritance.

Big Question: Why was Mendel's experimental work so successful?

Watch: The Work of Gregor Mendel

Johann Gregor Mendel (1822–1884) was a lifelong learner, teacher, scientist, and man of faith. As a young adult, he joined the Augustinian Abbey of St. Thomas in Brno in what is now the Czech Republic. Supported by the monastery, he taught physics, botany, and natural science courses at the secondary and university levels.

In 1856, he began a decade-long research pursuit involving inheritance patterns in honeybees and plants, ultimately settling on pea plants as his primary model system. (A model system is a system with convenient characteristics used to study a specific biological phenomenon to be applied to other systems.) In 1865, Mendel presented the results of his experiments with nearly 30,000 pea plants to the local Natural History Society. He demonstrated that traits are transmitted from parents to offspring independently of other traits and in dominant and recessive patterns. In 1866, he published his work in the proceedings of the Natural History Society of Brünn.

Mendel’s work went virtually unnoticed by the scientific community, which believed, incorrectly, that the process of inheritance involved a blending of parental traits that produced an intermediate physical appearance in offspring. The blending theory of inheritance asserted that the original parental traits were lost or absorbed by the blending in the offspring, but we now know that this is not the case.

This hypothetical process appeared to be correct because of what we know now as continuous variation. Continuous variation results from the action of many genes to determine a characteristic like human
height. Offspring appear to be a “blend” of their parents’ traits. For example, your skin, hair, or eye color might be a blend of traits passed down from both your mother and your father.

Instead of continuous characteristics, Mendel worked with traits that were inherited in distinct classes (specifically, violet versus white flowers); these types of traits are referred to as discontinuous variation. Mendel’s choice of these kinds of traits allowed him to see experimentally that the traits were not blended in the offspring, nor were they absorbed, but rather that they kept their distinctness and could be passed on.

In an interesting turn of events, in 1868, Mendel became abbot of the monastery and exchanged his scientific pursuits for his pastoral duties. He was not recognized for his extraordinary scientific contributions during his lifetime. In fact, it was not until 1900 that his work was rediscovered, reproduced, and revitalized by scientists on the brink of discovering the chromosomal basis of heredity.

Read: Mendel’s Experiments and Punnett Squares

Overview

Mendel’s seminal work was accomplished using the garden pea to study inheritance. This species naturally self-fertilizes, such that pollen encounters ova within individual flowers. The flower petals remain sealed tightly until after pollination, preventing pollination from other plants. The result is highly inbred, or “true-breeding,” pea plants. These are plants that always produce offspring that look like the parent. By experimenting with true-breeding pea plants, Mendel avoided the appearance of unexpected traits in offspring that might occur if the plants were not true-breeding. The garden pea also grows to maturity within one season, meaning that several generations could be evaluated over a relatively short time. Finally, large quantities of garden peas could be cultivated simultaneously, allowing Mendel to conclude that his results did not come about simply by chance.

Mendelian Crosses

Mendel performed **hybridizations**, which involve mating two true-breeding individuals that have different traits. In the pea, this is done by manually transferring pollen from the anther of a mature pea plant of one variety to the stigma of a separate mature pea plant of the second variety. In plants, pollen carries the male gametes (sperm) to the stigma, a sticky organ that traps pollen and allows the sperm to move down the pistil to the female gametes (ova) below. To prevent the pea plant that was receiving pollen from self-fertilizing and confounding his results, Mendel painstakingly removed all of the anthers from the plant’s flowers before they had a chance to mature.

Plants used in first-generation crosses were called **P0**, or parental generation one. After each cross, Mendel collected the seeds belonging to the P0 plants and grew them the following season. These offspring were called the **F1**, or the first filial (filial = offspring, daughter or son) generation. Once Mendel examined the characteristics in the F1 generation of plants, he allowed them to self-fertilize naturally. He then collected and grew the seeds from the F1 plants to produce the **F2**, or second filial, generation. Mendel’s experiments extended beyond the F2 generation to the F3 and F4 generations, and so on, but it was the ratio of characteristics in the P0–F1–F2 generations that were the most intriguing and became the basis for Mendel’s postulates.
In one of his experiments on inheritance patterns, Mendel crossed plants that were true-breeding for violet flower color with plants true-breeding for white flower color (the P generation). The resulting hybrids in the F1 generation all had violet flowers. In the F2 generation, approximately three quarters of the plants had violet flowers, and one quarter had white flowers.

Garden Pea Characteristics Revealed the Basics of Heredity

In his 1865 publication, Mendel reported the results of his crosses involving seven different characteristics, each with two contrasting traits. A trait is defined as a variation in the physical appearance of a heritable characteristic. The characteristics included plant height, seed texture, seed color, flower color, pea pod size, pea pod color, and flower position. For the characteristic of flower color, for example, the two contrasting traits were white versus violet. To fully examine each characteristic, Mendel generated large numbers of F1 and F2 plants, reporting results from 19,959 F2 plants alone. His findings were consistent.

First, Mendel confirmed that he had plants that bred true for white or violet flower color. Regardless of how many generations Mendel examined, all self-crossed offspring of parents with white flowers had white flowers, and all self-crossed offspring of parents with violet flowers had violet flowers. In addition, Mendel confirmed that, other than flower color, the pea plants were physically identical.
Once these validations were complete, Mendel applied the pollen from a plant with violet flowers to the stigma of a plant with white flowers. After gathering and sowing the seeds that resulted from this cross, Mendel found that 100 percent of the F1 hybrid generation had violet flowers. Conventional wisdom at that time would have predicted the hybrid flowers to be pale violet or for hybrid plants to have equal numbers of white and violet flowers. Mendel’s results demonstrated that the white flower trait in the F1 generation had completely disappeared.

Importantly, Mendel did not stop his experimentation there. He allowed the F1 plants to self-fertilize and found that, of F2-generation plants, 705 had violet flowers and 224 had white flowers. This was a ratio of 3.15 violet flowers per one white flower, or approximately 3:1. When Mendel transferred pollen from a plant with violet flowers to the stigma of a plant with white flowers and vice versa, he obtained about the same ratio regardless of which parent, male or female, contributed which trait. This is called a reciprocal cross — a paired cross in which the respective traits of the male and female in one cross become the respective traits of the female and male in the other cross. For the other six characteristics Mendel examined, the F1 and F2 generations behaved in the same way as they had for flower color. One of the two traits would disappear completely from the F1 generation only to reappear in the F2 generation at a ratio of approximately 3:1.

Upon compiling his results, Mendel concluded that the characteristics could be divided into expressed (dominant) and latent (recessive) traits. Dominant traits are those that are inherited unchanged in a hybridization. Recessive traits become latent, or disappear, in the offspring of a hybridization. The recessive trait does, however, reappear in the progeny of the hybrid offspring. An example of a dominant trait is the violet-flower trait. For this same characteristic, white-colored flowers are a recessive trait. The fact that the recessive trait reappeared in the F2 generation meant that the traits remained separate in the plants of the F1 generation. Mendel also proposed that plants possessed two copies of the trait for the flower-color characteristic, and that each parent transmitted one of its two copies to its offspring, where they came together.

Reflect Poll: A Decade of Research?

Gregor Mendel’s research was a decade-long pursuit. Why do you think he would spend a decade of his life researching pea plants?

- He joined the monastery and had plenty of time on his hands.
- He took his job of caring for the gardens at the monastery very seriously.
- He was a lifelong learner and simply curious.

Expand: Creating a Punnett Square

To understand the laws of probability used to predict the outcome of cross-breeding, you can visualize it by creating what is called a Punnett square. To prepare a Punnett square, all possible combinations of the parental alleles are listed along the top (for one parent) and side (for the other parent) of a grid, representing their meiotic segregation into haploid gametes. Then the combinations of egg and sperm are written in the table’s boxes to show which alleles are combining. Each box then represents the diploid genotype of a zygote, or fertilized egg, that could result from the specific mating. Because each possibility is equally likely, genotypic ratios can be determined from a Punnett square. If the pattern of inheritance is known, the phenotypic ratios can be inferred as well. For a monohybrid cross of two true-breeding parents, each parent contributes one type of allele. In the following example, only one genotype is possible. All offspring are Yy and have yellow seeds.
In this second example, the P generation, pea plants that are true-breeding for the dominant yellow phenotype, are crossed with plants with the recessive green phenotype. This cross produces F1 heterozygotes with a yellow phenotype. Punnett square analysis can be used to predict the genotypes of the F2 generation. A self-cross of one of the Yy heterozygous offspring can be represented in a 2 × 2 Punnett square because each parent can donate one of two different alleles. Therefore, the offspring can potentially have one of four allele combinations: YY, Yy, yY, or yy. Notice that there are two ways to obtain the Yy genotype: a Y from the egg and a y from the sperm, or a y from the egg and a Y from the sperm. Both of these possibilities must be counted.

Recall that Mendel’s pea-plant characteristics behaved in the same way in reciprocal crosses. Therefore, the two possible heterozygous combinations produce offspring that are genotypically and phenotypically identical despite their dominant and recessive alleles deriving from different parents. They are grouped together. Because fertilization is a random event, we expect each combination to be equally likely and for the offspring to exhibit a ratio of YY:Yy:yY:yy genotypes of 1:2:1. Furthermore, because the YY and Yy offspring have yellow seeds and are phenotypically identical, applying the sum rule of probability, we expect the offspring to exhibit a phenotypic ratio of 3 yellow:1 green. Indeed, working with large sample sizes, Mendel observed approximately this ratio in every F2 generation resulting from crosses for individual traits.
Lesson Toolbox

Additional Resources and Readings

An Amoeba Sisters video covering Punnett squares and genetic probabilities
  - Link to resource: https://www.youtube.com/watch?v=i-0rSv6oxSY&list=PLwL0Myd7Dk1Hj8WCDIDVBkqT-ZVdj7Js

A simulation of Mendel's pea plant experiment
  - Link to resource: https://scratch.mit.edu/projects/43261172/

An interactive pea experiment where you can breed your own hybrid pea plants
  - Link to resource: http://www.sonic.net/~nbs/projects/anthro201/exper/

A video that explains how Punnett squares work
  - Link to resource: https://www.youtube.com/watch?v=wAPpKtwOayk

Lesson Glossary

dominant: trait which confers the same physical appearance whether an individual has two copies of the trait or one copy of the dominant trait and one copy of the recessive trait
F1: first filial generation in a cross; the offspring of the parental generation
F2: second filial generation produced when F1 individuals are self-crossed or fertilized with each other
genotype: underlying genetic makeup, consisting of both physically visible and non-expressed alleles, of an organism
heterozygous: having two different alleles for a given gene on the homologous chromosome
hybridizations: process of mating two individuals that differ with the goal of achieving a certain characteristic in their offspring
P0: parental generation in a cross
phenotype: observable traits expressed by an organism
Punnett square: visual representation of a cross between two individuals in which the gametes of each individual are denoted along the top and side of a grid, respectively, and the possible zygotic genotypes are recombined at each box in the grid
recessive: trait that appears "latent" or non-expressed when the individual also carries a dominant trait for that same characteristic; when present as two identical copies, the recessive trait is expressed
reciprocal cross: paired cross in which the respective traits of the male and female in one cross become the respective traits of the female and male in the other cross
trait: variation in the physical appearance of a heritable characteristic

Check Your Knowledge

1. Mendel performed hybridizations by transferring pollen from the _______ of the male plant to the female ova.
   a. anther
   b. pistil
   c. stigma
   d. seed
2. A __________ is defined as a variation in the physical appearance of a heritable characteristic.
   a. trait
   b. continuous variation
   c. discontinuous variation
   d. model system

3. In pea plants, smooth seeds (S) are dominant to wrinkled seeds (s). In a genetic cross of two plants that are heterozygous for the seed shape trait, the Punnett square is shown. What is the missing genotype?
   a. SS
   b. Ss
   c. sS
   d. Ss

Answer Key:

Citations

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