

As you have learned, Mendel's monohybrid crosses were based on one characteristic controlled by one gene such as height (tall or dwarf) or seed shape (round or wrinkled) in a pea plant. As far as we currently know, genetic disorders such as cystic fibrosis and Huntington's disease each involve only one gene. What happens when more than one characteristic is involved in a cross? Gregor Mendel asked such a question and then conducted experiments to determine the answer.

Dihybrid Crosses and the Law of Independent Assortment

Mendel focused on two characteristics: seed shape and seed colour. Recall that the dominant allele for seed shape is round (R) while the recessive allele is wrinkled (r). The dominant allele for seed colour is yellow (Y), while the recessive allele is green (y). Mendel crossed two individuals that were heterozygous for seed shape (Rr) and seed colour (Yy).

A cross between two individuals for two pairs of heterozygous alleles is called a **dihybrid cross**. In this case the individuals were both $RrYy$. **Figure 1** shows how Mendel used true-breeding parent plants to produce plants (the F_1 generation) that were heterozygous for these two traits. He then used these F_1 plants to perform a dihybrid cross.

dihybrid cross a cross that involves two genes, each consisting of heterozygous alleles

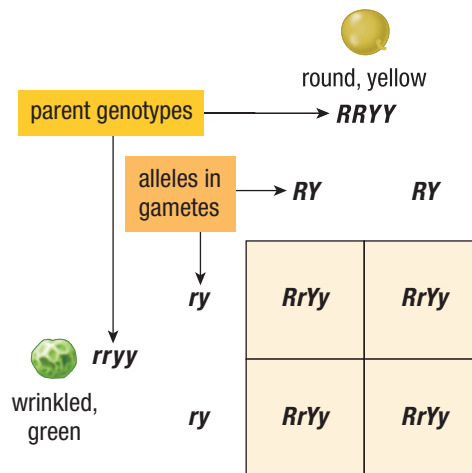


Figure 1 Mendel crossed parents that were homozygous for two traits to produce two pairs of heterozygous alleles for his dihybrid cross experiments.

A heterozygous individual for two characteristics will produce four possible gametes. A parent that is $RrYy$ can generate the possible gametes RY , Ry , rY , and ry . You will see in **Figure 2** that the alleles of the two genes— R and r , and Y and y —separate independently during the formation of the gametes. This is called Mendel's **law of independent assortment**. Each allele is independent of the other, and no two alleles are linked to each other. The alleles are found on different chromosomes, creating four different gametes in different combinations of the four alleles (R , r , Y , y).

law of independent assortment if genes are located on separate chromosomes, they will be inherited independently of one another

LEARNING TIP

Gene Order

When completing the Punnett square for a dihybrid cross, keep the genes in alphabetical order and write the dominant alleles (capital letters) first, for example, $GgRr$.

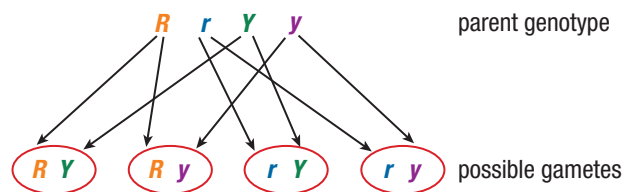


Figure 2 A pea plant that is heterozygous for both seed shape and seed colour can produce four different possible gametes. The four gametes encompass all the possible combinations. There are four possible combinations (RY , Ry , rY , and ry), and all are equally likely.

Mendel crossed numerous heterozygous F_1 generation plants. He then counted the F_2 offspring and noticed that the characteristics were *not* linked. He noted that although the original parents (or “grand-parents” of the F_2 generation) were either round yellow or wrinkled green, the F_2 offspring characteristics were of every combination possible, including round green and wrinkled yellow. It appeared that the inheritance of seed shape had no influence over the inheritance of seed colour. Four distinct combinations of seeds were produced in a 9:3:3:1 ratio (Figure 3). The alleles that control these two characteristics assort themselves independently.

cross: $RrYy \times RrYy$

gametes (pollen)

gametes (eggs)

















	<i>RY</i>	<i>Ry</i>	<i>rY</i>	<i>ry</i>
<i>RY</i>	 <i>RRYY</i>	 <i>RRYy</i>	 <i>RrYY</i>	 <i>RrYy</i>
<i>Ry</i>	 <i>RRYy</i>	 <i>RRyy</i>	 <i>RrYy</i>	 <i>Rryy</i>
<i>rY</i>	 <i>RrYY</i>	 <i>RrYy</i>	 <i>rrYY</i>	 <i>rrYy</i>
<i>ry</i>	 <i>RrYy</i>	 <i>Rryy</i>	 <i>rrYy</i>	 <i>rryy</i>

Figure 3 A summary of the dihybrid cross—9 round yellow seeds, 3 round green, 3 wrinkled yellow, and 1 wrinkled green. A dihybrid cross results in a phenotypic ratio of 9:3:3:1.

Tutorial 1 Solving Dihybrid Cross Problems

A dihybrid cross is an extension of a monohybrid cross. It involves two genes and up to four alleles.

Sample Problem 1: Determining Phenotypic Ratio

In watermelons, the green colour gene (G) is dominant over the striped colour gene (g), and round shape (R) is dominant over long shape (r). A heterozygous round green colour ($GgRr$) watermelon plant is crossed with another heterozygous round green colour ($GgRr$) plant. Determine the expected phenotypic ratio of the F_1 generation.

Step 1. Determine the possible gametes from each parent.

Each parent is heterozygous for green colour and round shape ($GgRr$). The alleles assort independently of each other. Four different gametes are produced (Figure 4).

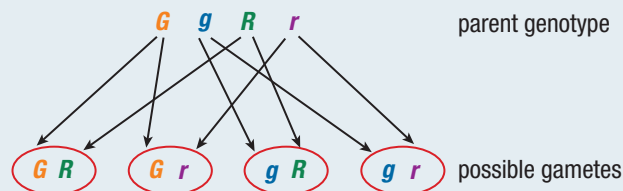


Figure 4

Step 2. Draw a Punnett square of the dihybrid cross.

Since there are four possible gametes from each parent, draw a 4×4 grid, with the possible gametes from one parent across the top and the possible gametes from the other parent down the left-hand side.

Step 3. Execute the cross.

Be careful when combining alleles. Work systematically across a row while keeping track of which four alleles will go into each square. Pair up the alleles for the same gene, as shown in Figure 5.

	<i>GR</i>	<i>Gr</i>	<i>gR</i>	<i>gr</i>
<i>GR</i>	$GGRR$	$GGRr$	$GgRR$	$GgRr$
<i>Gr</i>	$GGRr$	$GGrr$	$GgRr$	$Ggrr$
<i>gR</i>	$GgRR$	$GgRr$	$ggRR$	$ggRr$
<i>gr</i>	$GgRr$	$Ggrr$	$ggRr$	$ggrr$

Figure 5

Step 4. Compile a list of plants that have the same phenotypes.

Start with plants that will exhibit both dominant characteristics, followed by plants that are dominant for only one of the characteristics, and then finally with plants that are recessive for both characteristics.

green and round: $GGRR$, $GGRr$, $GgRR$, $GgRr$, $GGRr$, $GgRr$, $GgRR$, $GgRr$ (total = 9)

green and long: $GGrr$, $Ggrr$, $Ggrr$ (total = 3)

striped and round: $ggRR$, $ggRr$, $ggRr$ (total = 3)

striped and long: $ggrr$ (total = 1)

Therefore, the phenotype ratio is 9 : 3 : 3 : 1 for a dihybrid cross. 🌐

Sample Problem 2: Examining a Homozygous Cross

Assume that in guinea pigs, black fur (B) is dominant over white fur (b), and a rough coat (R) is dominant over a smooth coat (r). If a black, rough-furred guinea pig that is homozygous dominant for both traits ($BBRR$) is crossed with a white, smooth-furred guinea pig ($bbrr$), what are the expected phenotypes in a large litter?

Step 1. Determine what the gametes will be from each parent.

Parent 1 is homozygous dominant for black and rough fur ($BBRR$). The only gamete that this individual can produce is BR (Figure 6). Parent 2 is homozygous recessive for white colour and smooth fur ($rrbb$). The only gamete that this individual can produce is br (Figure 7).

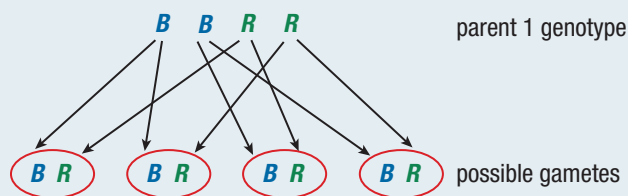


Figure 6

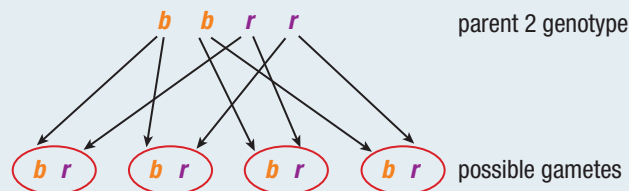


Figure 7

Step 2. Cross the gametes using a Punnett square.

Since only one type of gamete is supplied by each parent, only a 1×1 grid is needed (Figure 8).

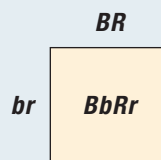


Figure 8

All of the offspring will be heterozygous for black and rough fur in any size litter.

Practice

1. In some breeds of dogs, a dominant allele controls the characteristic of barking (B) while on a scent trail. The allele for non-barking trailing dogs is (b). In these dogs an independent gene (E) produces erect ears and is dominant over drooping ears (e). For each of the following mating situations, calculate the phenotype ratio of the offspring:
 - (a) A non-barking trailer with erect ears (heterozygous) is mated with a heterozygous barking trailer with drooping ears ($bbEe \times Bbee$). [ans: non-barking trailer with erect ears: non-barking trailer with drooping ears: barking trailer with erect ears: barking trailer with drooping ears in a ratio of 1:1:1:1]
 - (b) A non-barking trailer with drooping ears is mated with a heterozygous barking trailer with drooping ears ($bbee \times Bbee$). [ans: non-barking trailers with drooping ears: barking trailer with drooping ears in a ratio of 1:1]
 - (c) A heterozygous barking trailer with heterozygous erect ears is mated with a heterozygous barking trailer with heterozygous erect ears ($BbEe \times BbEe$). [ans: barking trailer with erect ears: barking trailer with drooping ears: non-barking trailer with erect ears: non-barking trailer with drooping ears in a ratio of 9:3:3:1]
 - (d) A heterozygous barking trailer with heterozygous erect ears is mated with a non-barking trailer with drooping ears ($BbEe \times bbee$). [ans: non-barking trailer with erect ears: non-barking trailer with drooping ears: barking trailer with erect ears: barking trailer with drooping ears in a ratio of 1:1:1:1]

Probability

Genetic ratios are probabilities. Recall that if a cross occurs between two heterozygous individuals where one of the alleles is dominant, such as for widow's peak ($Ww \times Ww$), the phenotype ratio is 3:1. The total number of possible events is four, and the probability of producing an individual without a widow's peak (ww) is $\frac{1}{4}$, or 25 %. Similarly the probability of producing an individual with a widow's peak is $\frac{3}{4}$, or 75 %.

THE PRODUCT LAW

Many genetic events occur independently of one another. A couple has a 50 % chance of producing a boy for their first child. The probability of having a boy as a second child is also 50 %, because the sex of the second offspring is not affected by the sex of the first.

When two events are independent of each other, the probability that both events will occur can be calculated using the product law. The **product law** states that the probability of two or more outcomes occurring is equal to the product of their individual probabilities. The probability of giving birth to two boys is $\frac{1}{2} \times \frac{1}{2} = \frac{1}{4}$. Therefore, there is a 25 % chance of having two boys.

Recall that in a dihybrid cross the phenotypic ratio is 9:3:3:1. Consider Mendel's work with heterozygous yellow seeds crossed with heterozygous yellow seeds ($RrYy \times RrYy$). The probability of producing a round yellow seed is $\frac{9}{16}$ (about 56 %). You know that the alleles of seed colour and seed shape are independent of each other. If a cross is conducted between two heterozygous yellow seed plants (Yy), the probability of producing a yellow seed plant is $\frac{3}{4}$ (75%). If a cross is conducted between two heterozygous round seed plants, the probability of producing a round plant is $\frac{3}{4}$ (75%). Using the product rule, the probability of producing a round yellow seed plant from a cross between two heterozygous round yellow seed plants is $\frac{3}{4} \times \frac{3}{4} = \frac{9}{16}$.


The probability predicted by the product law is the same probability that is predicted by a dihybrid Punnett square. The product law also holds true for the other three probabilities in a dihybrid cross (**Table 1**). 

Table 1 Using the Product Law to Determine Probabilities of a Dihybrid Cross When Both Genes Are Heterozygous

Round or wrinkled seed: probability from monohybrid cross	Yellow or green seed: probability from monohybrid cross	Dihybrid cross probability
Round seed ($Rr \times Rr$) $\frac{3}{4} = 75\%$	Yellow seed ($Yy \times Yy$) $\frac{3}{4} = 75\%$	round, yellow seed ($RrYy \times RrYy$) $\frac{3}{4} \times \frac{3}{4} = \frac{9}{16}$ $75\% \times 75\% = 56.25\%$
round seed ($Rr \times Rr$) $\frac{3}{4} = 75\%$	green seed ($Yy \times Yy$) $\frac{1}{4} = 25\%$	round, green seed ($RrYy \times RrYy$) $\frac{3}{4} \times \frac{1}{4} = \frac{3}{16}$ $75\% \times 25\% = 18.75\%$
wrinkled seed ($Rr \times Rr$) $\frac{1}{4} = 25\%$	yellow seed ($Yy \times Yy$) $\frac{3}{4} = 75\%$	wrinkled, yellow seed ($RrYy \times RrYy$) $\frac{1}{4} \times \frac{3}{4} = \frac{3}{16}$ $25\% \times 75\% = 18.75\%$
wrinkled seed ($Rr \times Rr$) $\frac{1}{4} = 25\%$	green seed ($Yy \times Yy$) $\frac{1}{4} = 25\%$	round, green seed ($RrYy \times RrYy$) $\frac{1}{4} \times \frac{1}{4} = \frac{1}{16}$ $25\% \times 25\% = 6.25\%$

Discontinuous versus Continuous Variation

In Mendel's work with pea plants, the genes that control two characteristics did not interact with each other. This is the result of **discontinuous variation**. Pea plants were tall or short, and seeds were yellow or green. There were no in-between values.

product law the probability of two independent random events both occurring is the product of the individual probabilities of the events

CAREER LINK

Hybrid Research Manager

The managing of a hybrid breeding operation requires the ability to predict probable outcomes between different crosses. To learn more about careers in hybrid breeding operations,



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LEARNING TIP

Sample Size

Note that Mendel observed these ratios only because of his large sample size. He crossed hundreds of plants, producing thousands of offspring. As sample size increases, the experimental (actual) probability gets closer to the theoretical (expected) probability.

discontinuous variation when the expression of the products of one gene has no bearing on the expression of the products of a second gene

continuous variation when the product of one gene is affected by the product of another gene, the gene products may be additive, or one product may negate another product

additive allele an allele that has a partial influence on a phenotype

As you no doubt have noticed, there are many examples in nature where this is not the case. **Continuous variation** is when the phenotypic variation is not clear cut. For example, in the general population there are many variations of skin colour, from pale white to dark black. This is because skin colour is not controlled by one gene, but rather by three or more separately inherited genes from the father and mother. The genes are on different autosomal chromosomes and their interaction is additive. An **additive allele** contributes a set amount to a phenotype and is an example of continuous variation. Skin colour varies depending on which combination of six alleles the offspring inherit from parents. Each allele makes its own contribution and exhibits incomplete dominance.

Other alleles may also play a role. For example, alleles for freckles and red hair have a role in determining skin colour. Other traits that are under the control of additive alleles are height, hair colour, and eye colour. Continuous variation explains the substantial variation of phenotypes found in nature.

5.7 Summary

- Dihybrid crosses are crosses between individuals who differ in two pairs of alleles; if individuals are heterozygous for both alleles, the phenotype ratio of the offspring is 9:3:3:1.
- Mendel's law of independent assortment states that alleles of different genes separate into gametes independently of each other.
- The probability of two independent events both occurring may be calculated using the product law.
- Punnett square ratios are one way to show probability.
- Discontinuous variation occurs when a trait is either expressed or not. There is no in-between trait.
- Continuous variation occurs in nature when the expression of a characteristic is the sum of the expression of all alleles involved.

5.7 Questions

1. State Mendel's law of independent assortment. Provide an example that illustrates the law. **K/U**
2. List the possible gametes from the following individuals: **K/U**
 - (a) a widows-peaked (heterozygous), attached-earlobe individual
 - (b) a free-lobed (heterozygous), albino individual
 - (c) a dwarf, yellow seed colour (homozygous) pea plant
3. Assume that curly hair (*C*) is dominant to straight hair. Albinism (*P*) is recessive to normal skin pigmentation. A woman who is heterozygous for curly hair and albinism has a child. The father is homozygous dominant for curly hair and has albinism. **T/I**
 - (a) Determine the possible phenotypes for their child.
 - (b) Calculate the four different probabilities of a child being both a male and of each phenotype.
 - (c) What is the probability that the child will express albinism and have curly hair like his father?
4. Assume that in horses, black coat colour is dominant (*B*) to chestnut colour (*b*). The trotting gait is due to a dominant allele (*T*), and the pacing gait is a recessive allele (*t*). **T/I**
 - (a) If two black trotting horses are mated and have four offspring, all of which are black and pacing, what does that reveal about the probable genotypes of the parent horses?
 - (b) A stud is black and trotting. He is mated with a mare who is also black and trotting. Their colt is black and pacing. The same stud is mated to a second mare who is chestnut and pacing. Their colt is chestnut and trotting. What is the probable genotype(s) of the stud, mare 1, colt 1, mare 2, and colt 2?
5. In your own words, differentiate between continuous and discontinuous variation. **K/U**
6. Design an experiment in which you could determine whether height in tomato plants is governed by continuous variation or discontinuous variation. **T/I**
7. The alleles for human blood types A and B are codominant, but both are dominant over the type O allele. The Rh factor is separate from the ABO blood group and is located on a separate chromosome. The Rh+ allele is dominant over the Rh-. Indicate the possible phenotypes from mating a woman with type O, Rh-, with a man with type A, Rh+. The man is homozygous for type A and heterozygous for Rh factor. **T/I**
8. Why is sample size important in a genetic investigation? **A**
9. Is it possible to have a trihybrid cross? Explain your thinking. **T/I**