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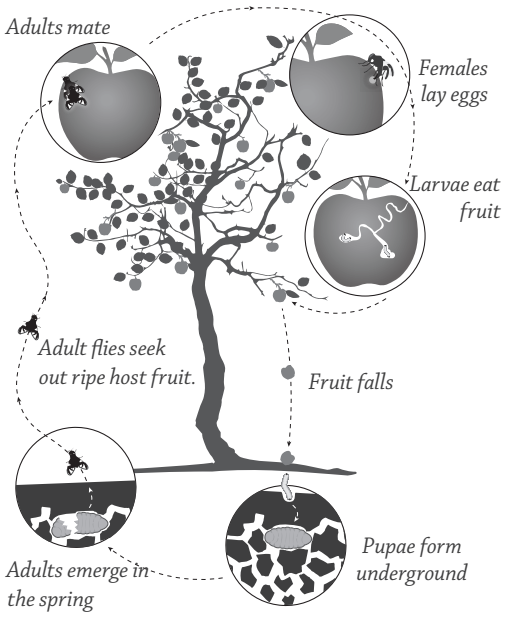
New Host, New Species? Host Fruit Preference

Guiding Question

Apples were brought to North America about 400 years ago. Sometime around 1850, some *Rhagoletis* flies moved from living on their native hawthorn fruit to living on apples. Is the population of hawthorn flies living on the apples becoming a new species?



Rhagoletis life cycle

Background

Adult *Rhagoletis* flies go to ripe fruit to find a mate and lay eggs. The offspring overwinter directly underneath the host tree they hatched in, and then emerge from the ground in the spring—and the cycle repeats.

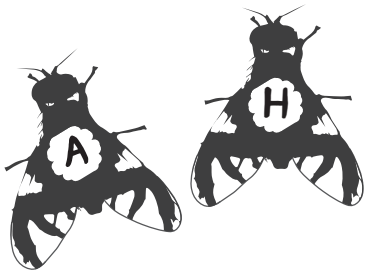
Experiment 1

Research question: When flies come out of the ground as adults, do they go back to the host fruit they hatched from, or instead randomly fly to either apples or hawthorns?

If the offspring from apple and hawthorn fly populations go back only to the same type of fruit they hatched from to mate and lay eggs, then the populations will be reproductively isolated.

Procedure

1. Catch adult flies as they emerge out of the ground under apple or hawthorn trees.
2. Mark each fly to show which host it came from. Release.
3. After a few days, re-capture flies from ripe fruit and count them.



Results *(data based on Feder et al, 1994)*

Host fruit that flies grew up in	Total re-captured	Host fruit where flies went to mate	
		Same fruit	Different fruit
Hawthorn fruit	45	43	2
Apple fruit	71	66	5

Question

1. In one sentence, summarize the results.

When flies come out of the ground as adults, they usually go back to the host fruit they hatched from.

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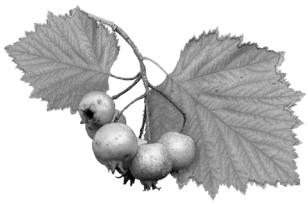
Experiment 2

Different types of host fruit have different odors. Odors from fruit are important signals that flies use to find a place to mate and lay eggs.

apple fruit



hawthorn fruit

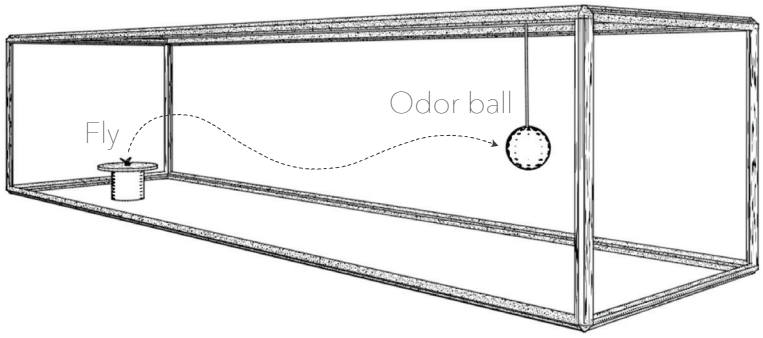
dogwood fruit

> **Research question:** Is fruit odor preference a reproductive barrier?

Using dogwood flies as a control (they are known to be a separate species), researchers compared fruit odor preferences among dogwood, apple, and hawthorn flies.

Procedure

1. In the fall, collect fruit with fly larvae living inside of them.
2. Take the fruit back to the lab and store each type of fly separately.
3. When flies emerge as adults in the spring, test them in a flight tunnel.
   a. To mimic ripe fruit, paint synthetic apple odors onto a red rubber ball.
   b. Place a fly onto a stand in the tunnel.
   c. Record whether the fly lands on the ball.



   d. Using a new fly each time, repeat until you have tested 100-200 flies. Repeat for hawthorn and dogwood odors. **Test each fly only once using only one of the three possible odors.**

Results *(data based on Dambroski, 2004)*

Population	Percent of flies that landed on the fruit bait		
	Hawthorn Odor	Apple Odor	Dogwood Odor
Hawthorn Flies	**77**	21	15
Apple Flies	15	**79**	11
Dogwood Flies*	21	13	**82**

* Dogwood flies are known to be a separate *Rhagoletis* species.
Note: No flies landed on a plain, odorless ball.

Questions

2. On the next page, plot the data from the table onto the bar graph. For each fly population, fill in 3 bars, one for each odor.

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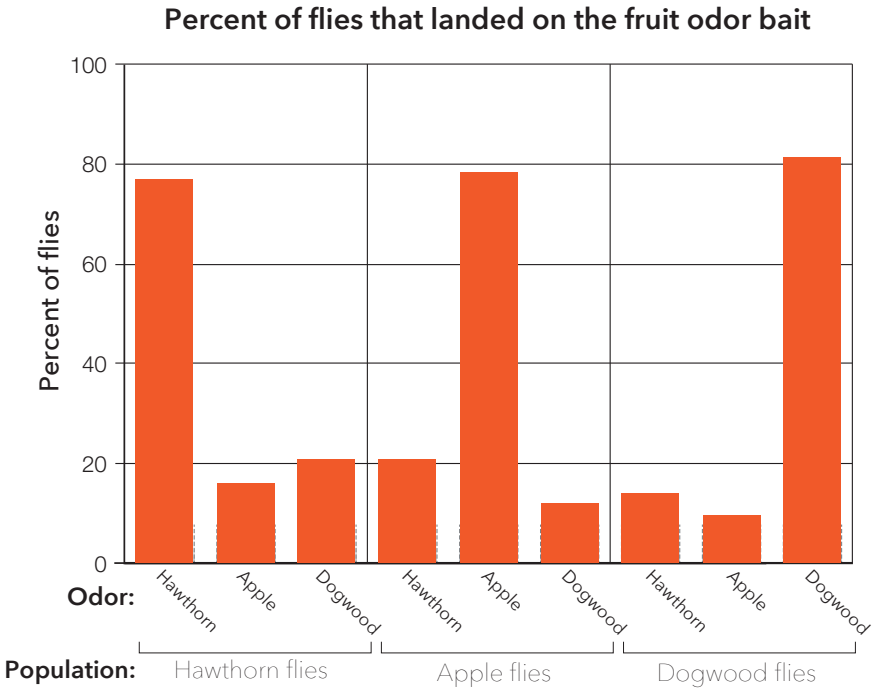

3. In one sentence, summarize the results.

Most flies are attracted to the odor of the fruit they grew up on, and a smaller number are attracted to other fruit odors.

Experiment 3:

According to one definition, two populations are considered to belong to the same species if they can produce fertile offspring together.

When researchers put hawthorn and apple flies together in the lab, they found that they could produce offspring together. These are referred to as hybrid offspring.

Research question: Which fruit odors are the hybrid offspring attracted to?

To find out, researchers tested the hybrid offspring in the same flight tunnel that they had used for experiment 2. For each odor, they tested over 100 flies.

Results *(data based on Lin et al, 2004)*

Population	Percent of flies that landed on bait	
	Hawthorn Odor	Apple Odor
Apple x Hawthorn hybrid	0	< 1

Questions

4. In one sentence, summarize the results.

Very few of the hybrid flies were attracted to either fruit odor.

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5. Compare and contrast the odor preferences of the hybrid offspring to those of the parent populations (from experiment 2).

Hybrid offspring fly to fruit odors far less often than their parents do. Parental flies must either be better at detecting fruit odors or more attracted to fruit odors than hybrid flies are.

6. Do you think that fruit preference is a reproductive barrier for the apple and hawthorn fly populations? Make a claim, and support it with evidence and reasoning from experiments 1 and 2.

Fruit preference is a reproductive barrier for apple and hawthorn fly populations.

Experiments 1 and 2 show that adult flies usually go back to the same type of fruit they grew up on. Since ripe fruit is where flies mate and lay eggs, they are most likely to mate with a fly of their same type.

7. Flies mate and lay their eggs on host fruit. In the wild, do you think hybrid offspring between apple and hawthorn flies are healthy, plentiful, and able to reproduce? Make a claim, and support it with evidence and reasoning from experiment 3.

Hybrid offspring are not healthy, plentiful, and able to reproduce.

Flies mate and lay their eggs on ripe fruit. Since they usually go back to the same fruit they grew up on, not very many hybrid offspring will be produced. And since hybrid flies are not attracted to fruit odor, they will probably not fly to ripe fruit and reproduce.

8. Attraction to fruit odor is a heritable trait influenced by genes. Do you think that differences in fruit odor attraction are being acted on by natural selection differently in apple vs. hawthorn flies? Make a claim, and support it with evidence and reasoning.

Differences in fruit odor attraction are being selected for in apple vs. hawthorn flies. Apple flies are most attracted to apple odors, and hawthorn flies are most attracted to hawthorn odors. Because attraction to fruit odor is important for reproduction, natural selection will favor flies that are attracted to good food sources.

References:

Dambroski, H. R. (2004). The role of diapause and host fruit odor preference in sympatric race formation of *Rhagoletis pomonella* (Doctoral dissertation). Retrieved from CurateND.

Lin, C.E., Dambroski, H.R., Feder, J.L., Berlocher, S.H., Nojima, S. & Roelofs, W.L. (2004). Postzygotic isolating factor in sympatric speciation in *Rhagoletis* flies: reduced response of hybrids to parental host-fruit odors. *Proceedings of the National Academy of Sciences of the United States of America*, 101(51), 17753-17758. doi: 10.1073/pnas.0408255101

NAME Answer key DATE

New Host, New Species? Life Cycle Timing

Guiding Question

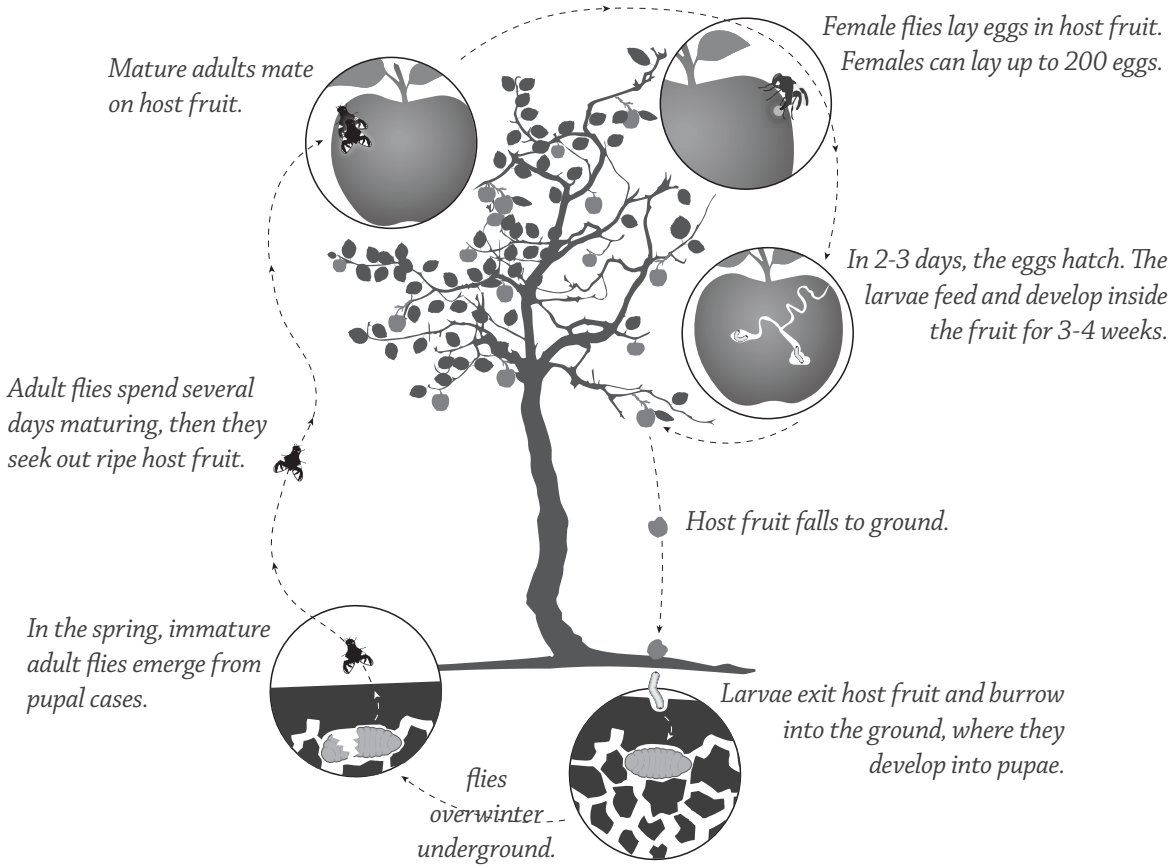
Apples were brought to North America, about 400 years ago. Sometime around 1850, some *Rhagoletis* flies moved from living on their native hawthorn fruit to living on apples. Is the population of hawthorn flies living on the apples becoming a new species?

Background

All species of *Rhagoletis* flies spend most of their lives underground and dormant. Adults emerge from the ground as their host fruit is getting ripe. They mature, mate, lay eggs, and die—all within a few weeks.

Timing is key to successful reproduction. Each species has a life cycle that is tied very closely to that of its host fruit. Dogwood fruit, for example, are ripe several days ***after*** hawthorn fruit—and flies from the dogwood species reach adulthood about 30 days ***later*** than hawthorn flies. Even though these two fly species live in the same area, adult flies exist at different times, and so they do not interbreed.

Could differences in life cycle timing also be a barrier to reproduction between apple and hawthorn fly populations?



Most *Rhagoletis* flies spend their whole lives very close to their host plant.

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Experiment 1

Adult flies mate on the fruit of their host plants, and they begin laying eggs in the fruit a short time later. For successful mating and egg laying, the fruit must be ripe enough for flies to smell it and soft enough for the female to puncture its skin. New groups of adult flies come out of the ground each day for several weeks. Each adult lives for about a month.

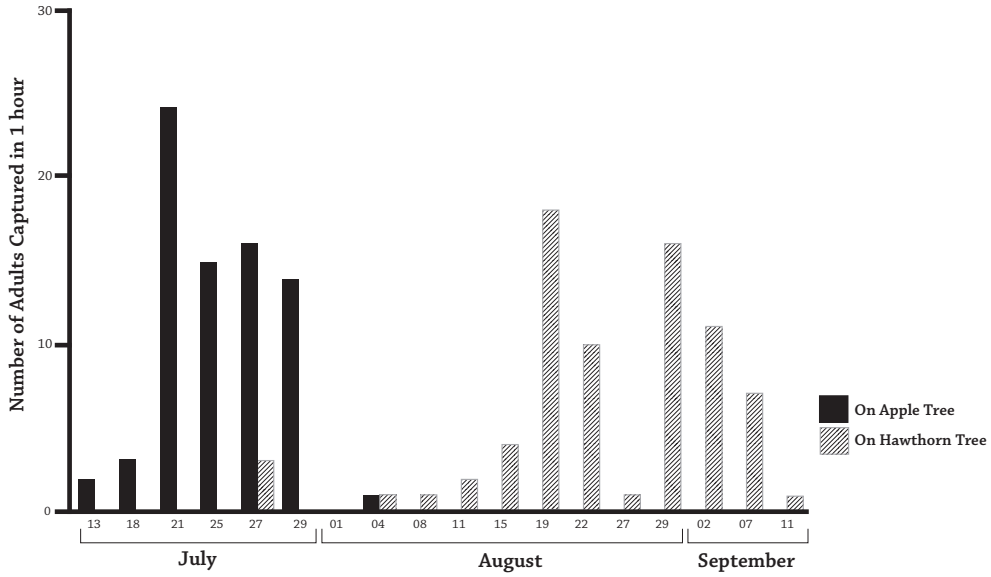
Research question: When are apple and hawthorn fruit ripe enough for flies to be mating on them?

Procedure

1. For one hour, capture and count flies that are mating on and laying eggs in hawthorn fruit. Repeat for apple fruit.

2. Repeat daily until no more flies are mating and laying eggs.

Results *(data based on Feder et al, 1993)*



Questions

1. In one sentence, summarize the results.

In general, flies mate and lay eggs on apples about a month earlier than they mate and lay eggs on hawthorns. (or -- Apples are ripe about a month earlier than hawthorn fruit.)

2. Given that adult flies live for only a few weeks after they begin mating, do you think the same flies are mating on both apple and hawthorn fruit? Explain.

No. Most of the flies that mate on apples probably die before hawthorn fruit is ripe.

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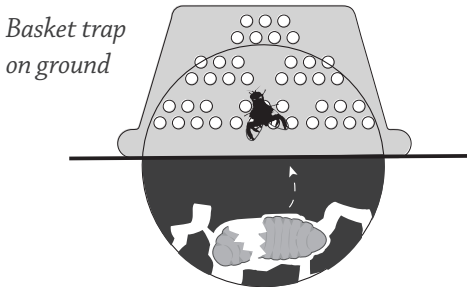
Experiment 2

Research question: When do apple and hawthorn fly adults emerge from the ground? How does this compare to when fruit are ripe?

Since fly larvae crawl out of fallen fruit and directly into the ground, adults emerge directly below the tree where its parents mated and laid eggs.

Procedure

1. Shortly before adult flies begin to emerge, place fly traps over the ground directly below apple and hawthorn trees.
2. Every week, empty the traps and count the flies.
3. When no more adults are emerging, add up the total numbers of apple and hawthorn flies that emerged, and calculate the percentage of flies that emerged each week.

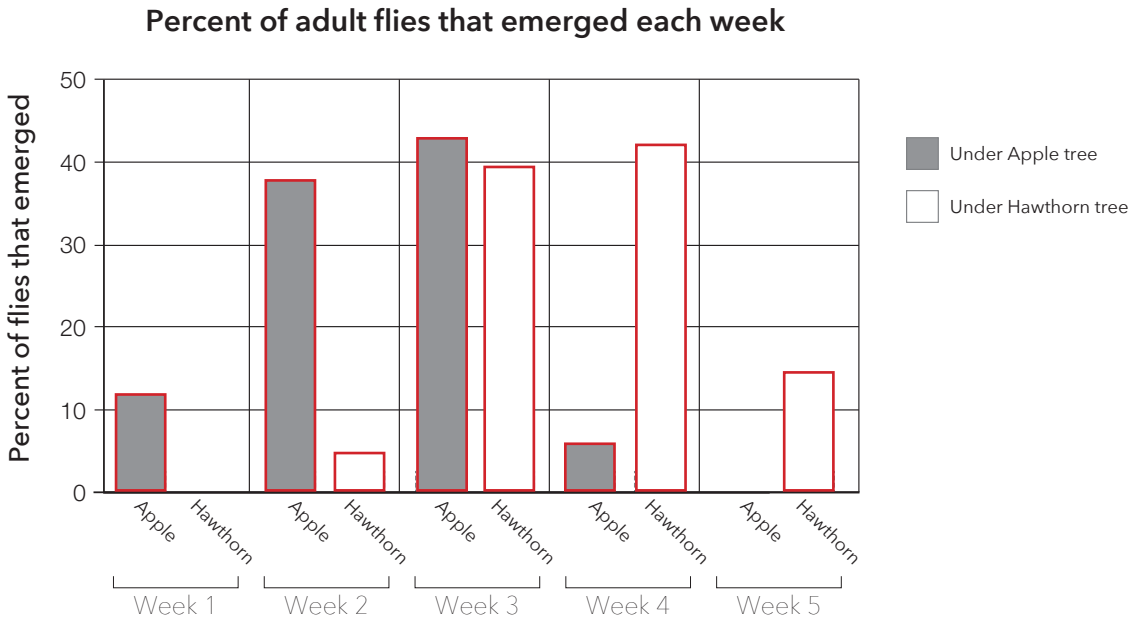


Results *(data based on Feder et al, 1993)*

Week	Dates	Percent of adult flies that emerged each week	
		Under Apple tree	Under Hawthorn tree
1	June 25 – July 1	12	0
2	July 2 – 8	38	4
3	July 9 – 15	43	39
4	July 16 – 22	7	42
5	July 23 – 29	0	15

Questions

3. Using the data from the table above, add bars to the graph:



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4. In one sentence, summarize the results.

Flies emerge over several days, and apple flies tend to emerge earlier than hawthorn flies.

Experiment 3

In experiment 2, there were many variables that researchers could not control. For example, maybe there was a difference in the nutrients or temperature of the soil that was affecting when the flies emerged. They decided to test emergence time in the lab to see if the results were the same.

Research question: Is earlier emergence time a heritable trait that could be selected for in the apple fly population?

Procedure

1. In the fall, collect apple and hawthorn fruit with fly larvae inside of them.
2. Take the fruit to the lab and store each type of fly separately. Wait for them to become pupae.
3. To mimic winter, keep the pupae cold for several months. To mimic spring, warm them.
4. Wait for adults to begin to emerge.
5. Every day, collect and count the newly emerged adults.

Results *(data based on Feder et al, 1993)*

Population	Number of pupae	Average emergence time of adults (number of days after pupae were warmed)
Larvae from apples	268	53
Larvae from hawthorns	106	61

Questions

5. In one sentence, summarize the results.

Like they did in the wild, the apple fly population emerged earlier than the hawthorn fly population.

6. Flies from apple and hawthorn fruit were raised in exactly the same conditions in the lab. Do the data from experiment 3 suggest that emergence time is environmentally driven, or is it more likely a heritable trait?

Emergence time is likely heritable.

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7. Data from other experiments (not shown) show that apple flies emerge from the ground and immediately begin to mate and lay eggs on ripe fruit. In contrast, hawthorn flies begin to mate and lay eggs on ripe fruit about 7-10 days after they emerge.

Do you think life cycle timing could be a barrier to reproduction between apple and hawthorn fly populations? Make a claim, and support it with evidence and reasoning.

Life cycle timing is starting to be a barrier to reproduction between apple and hawthorn fly populations. Because apple fly adults emerge earlier than hawthorn flies and begin to mate and lay eggs right away, they are more likely to mate and lay eggs on apple fruit, which are ripe earlier than hawthorn fruit.

8. Do you think that different heritable traits are being selected for in apple vs. hawthorn fly populations? Make a claim, and support it with evidence and reasoning.

Earlier emergence time is probably being selected for in the apple fly population, but not in the hawthorn fly population. Because apples are ripe before hawthorn fruit, any flies that emerge earlier would be more likely to mate and lay their eggs on apple fruit. They would then pass early emergence alleles to their offspring.

Reference
Feder, J.L., Hunt, T.A., & Bush, G. L. (1993). The effects of climate, host phenology and host fidelity on the genetics of apple and hawthorn infesting races of *Rhagoletis pomonella*. *Entomologia Experimentalis et Applicata, 69*(2), 117-135. doi: 10.1111/j.1570-7458.1993.tb01735.x

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New Host, New Species? Alleles

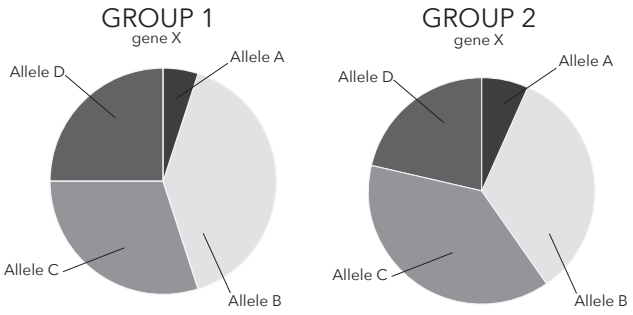
Guiding Question

Apples were brought to North America, about 400 years ago. Sometime around 1850, some *Rhagoletis* flies moved from living on their native hawthorn fruit to living on apples. Is the population of hawthorn flies living on the apples becoming a new species?

Background

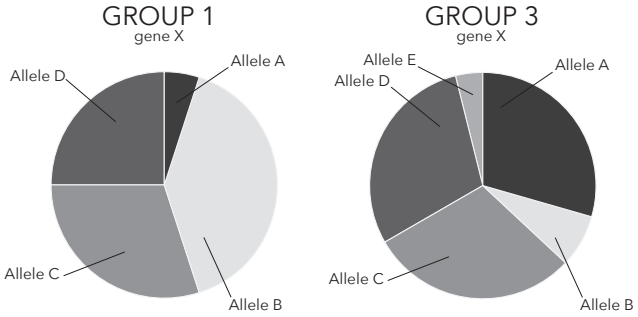
If flies from the apple and hawthorn populations are freely interbreeding with one another, then they would be considered the same species. One way we can tell if interbreeding is happening or not is to see if alleles (different versions of the same genes) in the populations are present in similar or different frequencies.

Group 1 and 2 are Interbreeding



If two groups are interbreeding freely, then alleles from the parents are mixed together in the offspring. We would expect to see the same alleles in both populations at about the same frequency.

Group 1 and 3 are NOT Interbreeding



If two groups are not interbreeding freely, then their alleles are ***not*** mixing in offspring. As natural selection causes reproductively isolated groups to become more genetically different over time, the allele frequencies shift differently in the two groups. Often, one group has alleles that another lacks completely.

Experiment 1

Research question: Do fly populations from apple vs. hawthorn fruit have different allele frequencies for some genes?

Procedure

1. Collect several hundred flies from hawthorn and apple fruit.
2. Isolated the flies' DNA. For several genes, determine what alleles each individual fly has.
3. For each population, calculated the allele frequencies: For a certain gene, what percentage of the alleles are allele A vs. allele B, etc.

Results are summarized in the tables on the next page.

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Allele Frequency Pie Charts

Worksheet

Instructions

Use the data from Experiment 1 (provided below) to fill in the pie charts. Color in the allele frequencies in both fly populations for each gene.

Each pie wedge represents 10%.

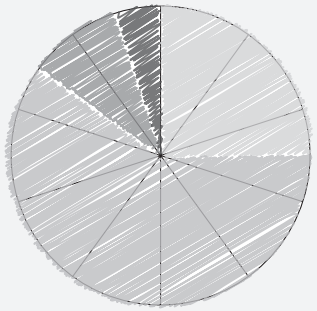
Use a different color for each allele. Mark the corresponding allele color in the circle on the bottom of the table.

EXAMPLE GENE

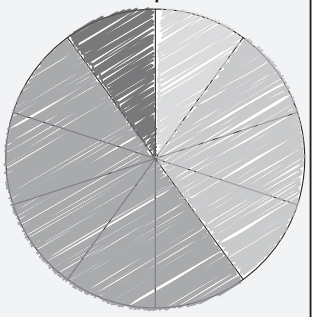
Allele Frequencies (%):

	Allele			
	A	B	C	D
Apple	25	60	10	5
Hawthorn	10	30	50	10

Apple Population Flies



Hawthorn Population Flies

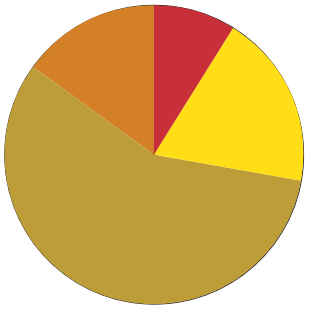


GENE 1

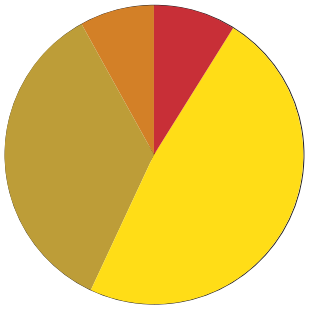
Allele Frequencies (%):

	Allele			
	A	B	C	D
Apple	9	19	57	15
Hawthorn	9	48	35	8

Apple Population Flies



Hawthorn Population Flies

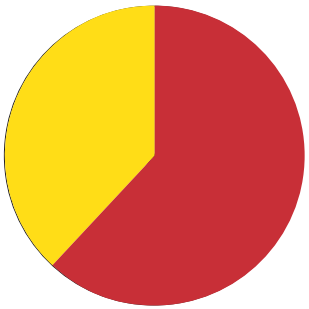


GENE 2

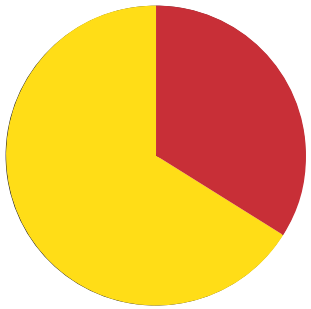
Allele Frequencies (%):

	Allele	
	A	B
Apple	62	38
Hawthorn	34	66

Apple Population Flies



Hawthorn Population Flies

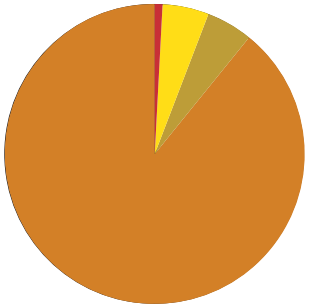


GENE 3

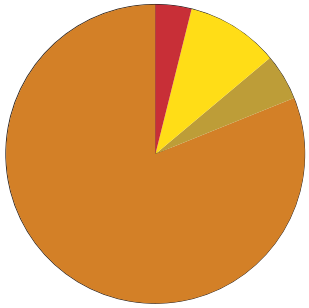
Allele Frequencies (%):

	Allele			
	A	B	C	D
Apple	1	5	5	89
Hawthorn	4	10	5	81

Apple Population Flies



Hawthorn Population Flies

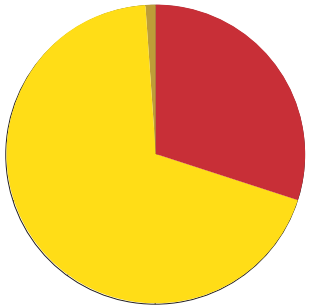


GENE 4

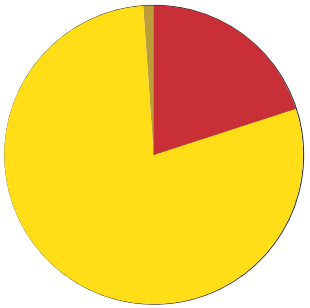
Allele Frequencies (%):

	Allele		
	A	B	C
Apple	30	69	1
Hawthorn	20	79	1

Apple Population Flies



Hawthorn Population Flies



(data based on Feder et al, 1990)

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Questions

1. Using the data in the tables on the Allele Frequency Pie Charts (page 2), color in the allele frequencies on the pie charts.

2. In one sentence, summarize the results.

The allele frequencies for genes 3 and 4 are similar between apple and hawthorn fly populations, but the frequencies for genes 1 and 2 are very different.

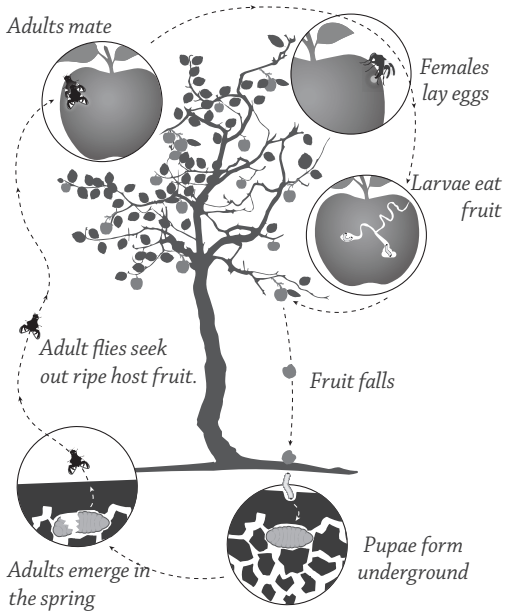
Experiment 2

In order for flies to mate and lay eggs, their life cycle must be timed so that they become adults when their host fruit is ripe. If they are too early or too late, they will miss their window of opportunity.

Apples get ripe about 30 days earlier than hawthorn fruit, and hawthorn flies vary in their emergence time. Researchers hypothesized that, after apples were introduced, hawthorn flies that became adults earlier were more likely to mate and lay eggs on apples.

Their logic looked like this:

- The timing of events in a fly's life cycle (like when adults emerge from the ground) is influenced by genes.
- Variations in these genes—in other words, alleles—may cause differences in life cycle timing.



Rhagoletis life cycle

- Alleles associated with an earlier emergence and mating time might have been acted upon by natural selection in the population of flies that moved from hawthorns to apples.

Research question: Are any alleles in hawthorn flies associated with differences in life cycle timing?

Procedure

1. Collect fly larvae from hawthorn fruit. Bring them to the lab and wait for them to become pupae.
2. To mimic winter, keep the pupae cold for several months. To mimic spring, warm them.
3. Wait for adults to begin to emerge.
4. Every day, collect the newly emerged adults.
5. Test the flies' DNA to see what alleles they have for genes 1–4, and calculate allele frequencies (for a certain gene, the percent of alleles that are A vs. B, etc.)

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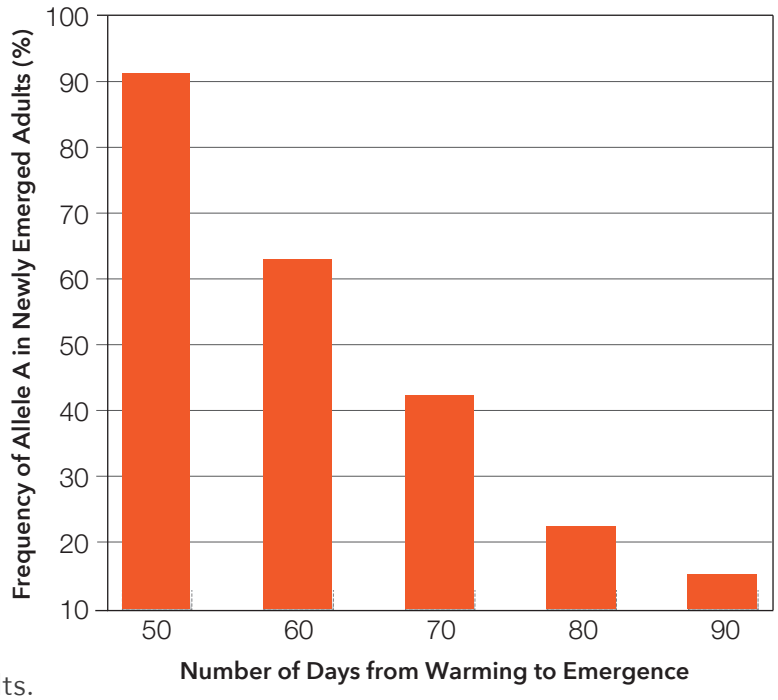
Results for Gene 2, Allele A

(Data based on Feder et al, 1993):

Days from warming to adults emerging*	Frequency of allele A (percent)
50	91
60	64
70	42
80	23
90	15

** Adult flies normally emerge from the ground at different times.*

Emergence Time vs. Allele Frequency for Gene 2



Questions

3. Use the data from the table to fill in the bar graph on the right.

4. In one sentence, summarize the results.

They frequency of allele A was highest in the hawthorn flies that emerged earliest, and lowest in the flies that emerged latest.

5. Compare the results above with those for Gene 2 in Experiment 1. Do the data support the researchers' hypothesis that hawthorn flies that became adults earlier were more likely to mate and lay eggs on apples? Explain.

Yes. Gene 2, Allele A is most frequent in hawthorn flies that emerge earlier, and also in apple flies. Since apples are ripe earlier than hawthorn fruit, the data support the hypothesis.

6. Do you think that alleles are freely mixing between apple and hawthorn fly populations? Make a claim, and support it with evidence and reasoning from experiment 1.

Alleles are not freely mixing between apple and hawthorn fly populations. The allele frequencies for genes 1 and 2 are very different between the two populations.

7. Do you think that different heritable traits are being selected for in the apple and hawthorn fly populations? Make a claim, and support it with evidence and reasoning from experiments 1 & 2.

Different heritable traits are being selected for in the apple and hawthorn fly populations. Earlier emergence time is a heritable trait (associated with Gene 2, Allele A), and natural selection has caused it to become more frequent in the apple fly population.

References

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