

IB BIOLOGY HIGHER LEVEL

Extended Essay

An analysis on the mode of inheritance and role of fungiform papillae in the bitter taste perception of Brassica Genus plants and Phenylthiocarbamide (PTC) strips

“How can bitter taste perception be explained in terms of fungiform papillae density and Mendelian genetics, and is the gene frequency of bitter taste perception in Hardy-Weinberg equilibrium?”

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Introduction

Background knowledge

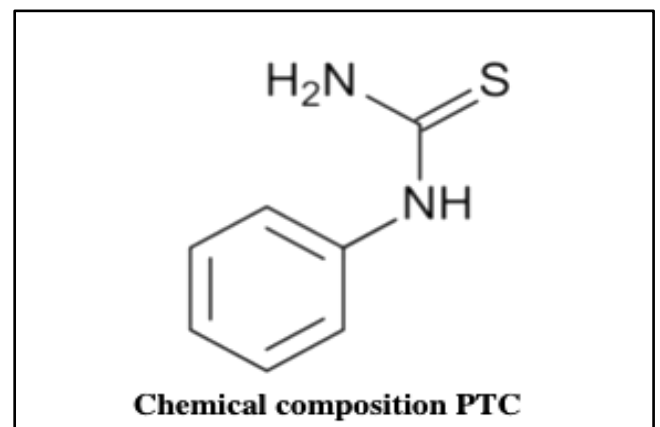
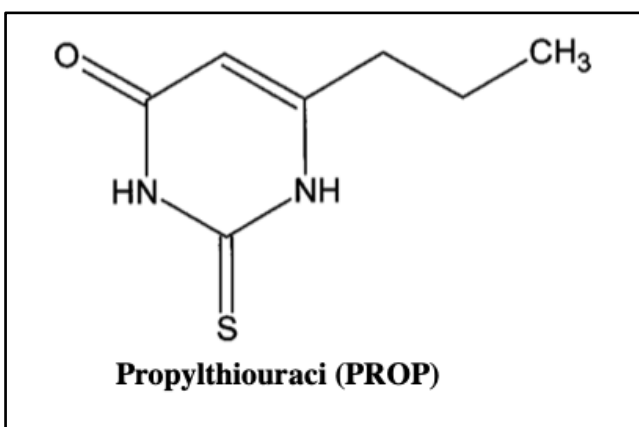
It is said there are five major taste modalities that most humans are able to taste; bitter, sweet, sour, salty and umami. Taste perception is an essential trait for the survival in the interplay between organisms and the environment, to help detect chemical substances contained in food, bitter taste sensitivity has also been shown to be important in food selection affecting individual's metabolism and physiology (Ruqaiya Hussain, 2014). It is thought to guard against consumption of toxins, many of which are bitter to humans (Melis and Tomassini Barbarossa, I., 2017). Almost 90 years ago, scientist A.L Fox poured a powdered form of phenylthiocarbamide (PTC) into a bottle when some of the powder accidentally blew into the air and another occupant of the laboratory complained of a bitter taste sensation. Curious to understand why they tasted this chemical substance differently a number of studies were conducted on families and different population samples.

Bitterness is a taste sensation that arises when precise chemical substances come into contact with specific receptors in specialized cells found on the human tongue (Smail, 2019).

Fungiform papillae are raised lingual structures located on the anterior two-thirds of the tongue, which play a vital role in human taste perception. These small bumps are made of several receptor cells all responsible for sensing a single flavor, the number of bumps present can vary from person to person. Geneticists have found inherent components that determine how people taste bitterness, it has been suggested that the ability to taste chemicals such as PTC is inherited in a classic Mendelian recessive manner. Alleles in the taste receptor membrane TASR38 is located on chromosome 7, this gene encodes for seven-transmembrane G protein-coupled receptors controlling for the ability to taste glucosinolates a family of

bitter tasting compounds found in Brassica Genus plants, phenylthiocarbamide and 6-n-propylthiouracil (PROP) (TASR38 taste 2 receptor membrane 38 - Gene - GTR - NCBI, 2020). Although several allelic forms of the TASR38 gene have been identified geneticists have discovered that there are two predominant forms responsible for expressing this homozygous recessive phenotype **T** and **t**, where **T** is the dominant tasting allele and **t** the recessive non-tasting allele.

Since the TASR38 taster gene is said to be dominant, an individual can still have the ability to taste prop-like chemicals, whether they carry two copies of the dominant gene or one. This explains the large variation in bitter taste intensity and perception among individuals, where some have the ability to taste glucosinolates, a family of bitter tasting compounds found in brassica genus plants and PTC. This ability to taste bitterness in certain chemicals like PTC is often referred to as the supertaster phenomenon. Supertasters are said to be more sensitive to certain flavors than the rest of us. This concept was first described by A.L Fox in 1932, when it was noted that PTC was tasteless to some individuals including himself, yet intensely bitter to others.



Significance and worthiness of experiment (Personal engagement)

Genetics has been an interest of mine ever since we began studying this as part of my IB Biology course. From the sight of articles and literature reviews on the topic of genetics and bitter taste perception, I was quickly determined to further investigate this phenomenon. I wanted to investigate the role of fungiform papillae density on bitter taste sensitivity determining whether there is a correlation between bitter taste intensity and fungiform papillae density. I also wanted to investigate the mode of inheritance of this phenotype confirming or rejecting a classic Mendelian manner, i.e. seeing whether it follows the 3:1 ratio, as well as determining whether my tested population followed the Hardy-Weinberg equilibrium principle.

Research question

“How can bitter taste perception be explained in terms of fungiform papillae density, Mendelian genetics, and is the gene frequency of bitter taste perception in Hardy-Weinberg equilibrium?”

Investigation

Aim of experiment

To answer the research, question the following aims have been set:

- Determine the relationship between fungiform papillae density and bitter taste sensitivity of different chemical substances (PTC and brassica genus plants)
- Conduct Chi-square tests to confirm or reject whether this bitter tasting phenotype follows a classic Mendelian ratio of 3:1. Further construct pedigree charts and Punnett squares to predict possible bitter tasting alleles in offspring within families.

- Calculating the gene frequencies of the tested population to determine whether they follow the Hardy-Weinberg equilibrium

Materials

- Brassica Genus plants (broccoli and Brussel sprouts)
- Phenylthiocarbamide strips (PTC)
- Blue food coloring (For staining tongue to allow clear counting of fungiform papillae)
- Cotton q-tips (for applying food coloring)
- Camera/phone for (For taking photos of tongue receptors)
- Hole punched paper (3.5 cm)
- Bitter taste rating scale (Refer to appendix I)
- Signed consent forms (Refer to appendix II and III)
- 8 families and 23 volunteers (ages 17-18)

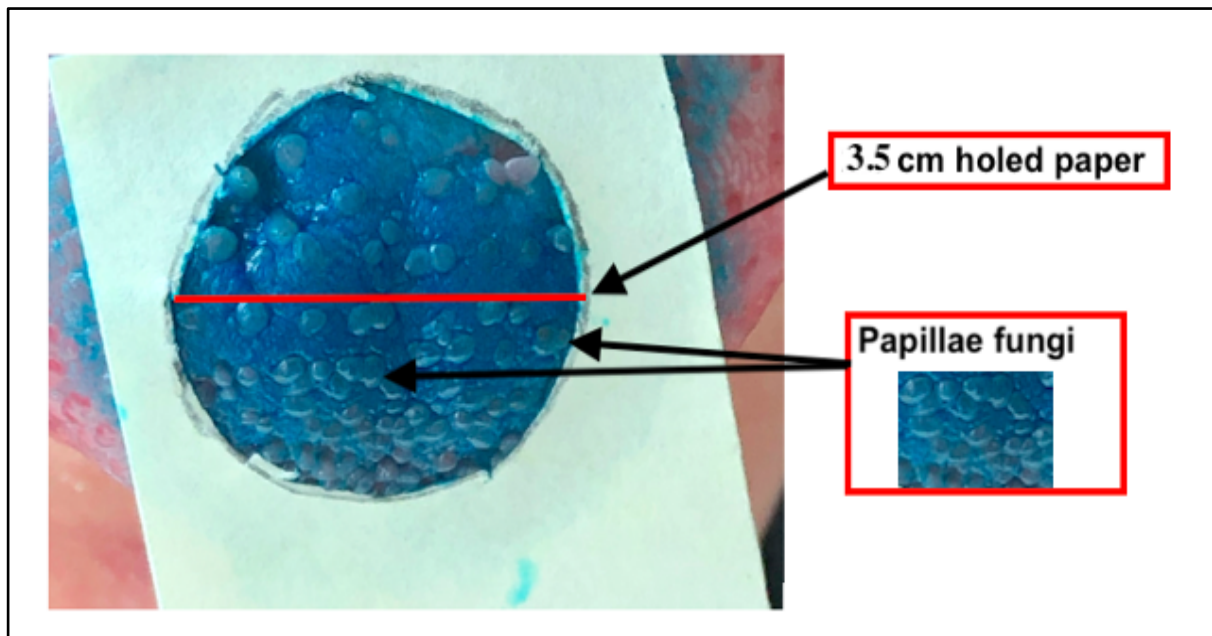
Experimental method

1. Participants were provided with the required materials; they were then instructed to taste the PTC strips and rate the bitterness on a given scale from 1-5 (Refer to appendix I)
2. They were then asked to wait for a short period of time before trying the brassica genus plants (broccoli and Brussel sprouts) to avoid any interfering tastes. Similarly, they were instructed to rate the bitter taste intensity
3. After tasting all substances, they were instructed to stain their tongues with blue food coloring using a q-tip. This would allow the color to slide off the prominent pink fungiform papillae and allow for easier counting. With the 3.5 cm hole punched paper they were instructed to place the paper on either the left or right counterpart of their

tongue (ensuring that every participant placed the paper around the same area of their tongue). A photo of their tongue was then taken to allow for fungiform papillae counting.

Similar procedure was used when assessing families, however the families only tasted PTC strips and I did not look into their fungiform papillae density, therefore step 2 or 3 was not followed. The results collected from families would allow me to further formulate and check the same principle only using PTC strips.

Diagram of procedure for counting fungiform papillae density



Method of calculating fungiform papillae density:

- Fungiform papillae were counted within the 3.5 cm area hole
- Formula used for calculating the density:

$$(\text{Number of counted fungiform papillae}) \div \pi (3.5\text{cm} \div 2)^2$$

Risk assessment

All chemicals used had no potential harm towards the participants, however it was important to ensure that no one was allergic to any of the substances before conducting the experiment.

Further, health concerns were considered during the experimental procedure due to Covid-19. The experiments were carefully conducted by each participant individually, they were all given instructions regarding the experiments as well as video and picture demonstrations. I was in no contact with any of the individuals during this process.

Hypothesis

We expect to see a strong correlation between the fungiform papillae density and bitter taste sensitivity rating of the different substances tasted. The larger the fungiform papillae density the higher the bitter taste intensity will be. It is also expected that the genotypic frequency follows a classic Mendelian manner showing a calculated ratio of 3:1. Lastly, it is expected that the population's genotype frequencies will be in Hardy-Weinberg equilibrium.

Variables

Independent variable

Source of bitter taste: The brassica genus plants (broccoli and Brussel sprouts) and Phenylthiocarbamide (PTC) strips

Dependent variable

The bitter taste intensity of each individual/family member (marked on indicated scale from 1-5)

Controlled variables

Controlled variable	Why it must be controlled	How it will be controlled
1. The bitterness rating scale	<ul style="list-style-type: none"> Ensures accurate results Rating of bitterness kept constant for everyone 	<ul style="list-style-type: none"> Same scale will be used for all participants to rate the bitterness (Refer to appendix I)
2. The age group of participants (Not controlled for family analysis)	<ul style="list-style-type: none"> It is Important to collect volunteers within same age range as this could impact results; perception may vary within different age groups 	<ul style="list-style-type: none"> Collect volunteers within the age range of 17-18 years old
3. Related family members	<ul style="list-style-type: none"> All families tested have to be related as I am analyzing the mode of inheritance Data will not be sufficient if family members are not related 	<ul style="list-style-type: none"> Families will be informed of this before partaking in the experiment
4. The different brassica plants tasted	<ul style="list-style-type: none"> Important that same type of vegetables are given to all individuals tested ensuring more accurate and sufficient results 	<ul style="list-style-type: none"> I will purchase the same vegetables and ensure everyone tastes the same one
5. The area on tongue where the fungiform papillae will be counted	<ul style="list-style-type: none"> This will allow more sufficient results if volunteers are instructed to place the holed paper on the same area of tongue 	<ul style="list-style-type: none"> Instruct the volunteers to palace the holed paper on the left or right counterpart of their tongue I will show demonstration videos to make this clear before the experiment
6. The area of the hole on the paper has to be 2.5 cm	<ul style="list-style-type: none"> This needs to be controlled as it will affect the calculated fungiform papillae density 	<ul style="list-style-type: none"> I will be cutting out 23 holed punched paper with the same diameter of 2.5 cm
7. Eating before the experiment	<ul style="list-style-type: none"> This is important to ensure there are no interfering flavors when the volunteers are tasting the different substances 	<ul style="list-style-type: none"> The volunteers will be instructed to avoid eating before the experiment

Results

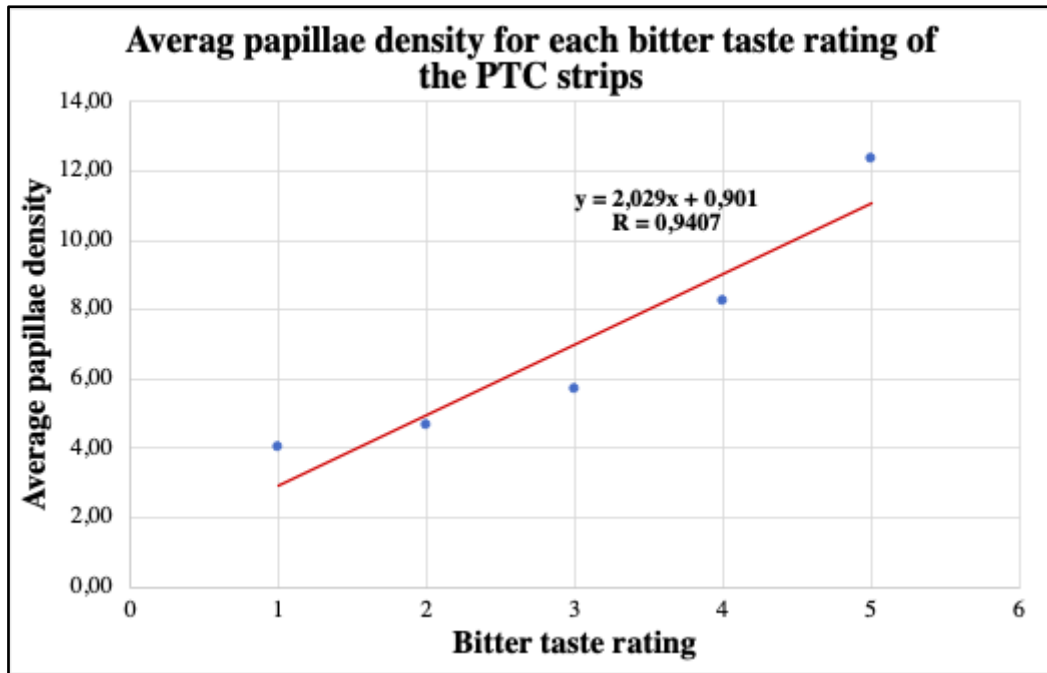
Figure I. Raw data table of collected results from volunteers

Participant	Number of counted fungiform papillae	Calculated density of fungiform papillae (per cm²)	PTC strips (Bitterness rating)	Broccoli (Bitterness rating)	Brussel sprouts (bitterness rating)
1	89	9,80	4	3	2
2	55	6,06	4	3	3
3	116	12,78	5	4	3
4	51	5,62	3	2	2
5	114	12,56	5	3	3
6	72	7,93	4	3	3
7	48	5,29	3	2	2
8	60	6,61	4	3	2
9	44	4,85	2	1	1
10	61	6,72	4	2	2
11	95	10,46	4	3	3
12	55	6,06	3	2	2
13	105	11,56	4	5	3
14	38	4,19	1	2	1
15	35	3,85	1	1	1
16	49	5,40	3	3	2
17	56	6,17	3	2	2
18	98	10,79	5	4	2
19	101	11,12	4	2	2
20	53	5,84	4	3	2
21	121	13,33	5	4	3
22	40	4,41	2	3	2
23	55	6,06	4	4	3

Correlation between density and bitter taste rating

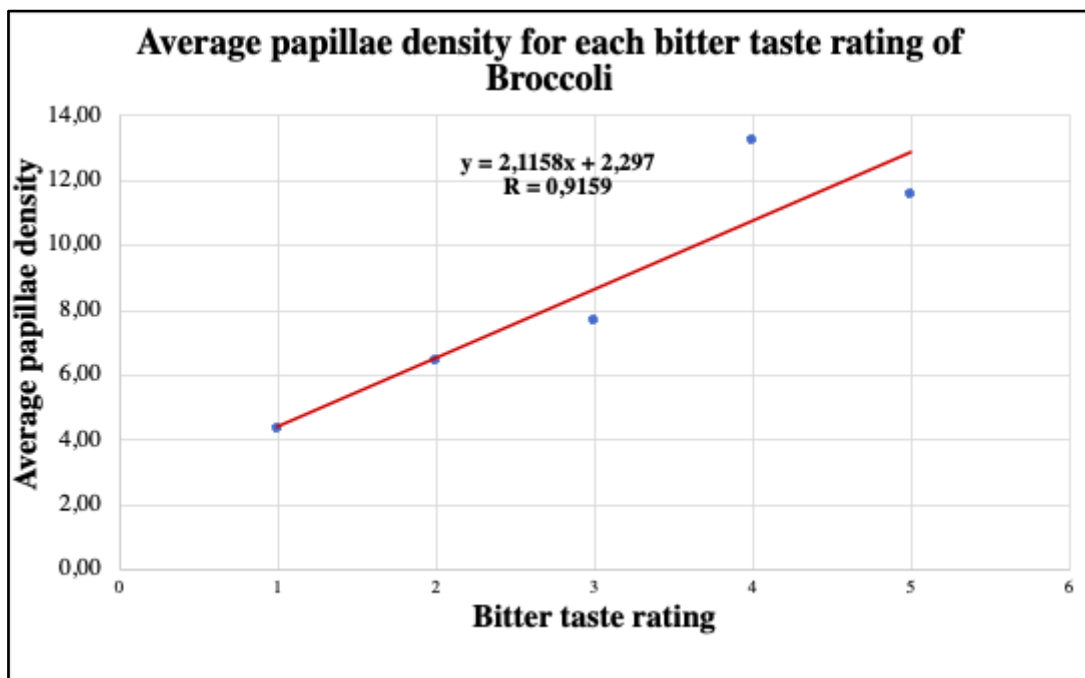
1. PTC Strips

Figure II. Linear graph displaying the relationship between bitter taste rating and average tongue papillae density for PTC strips



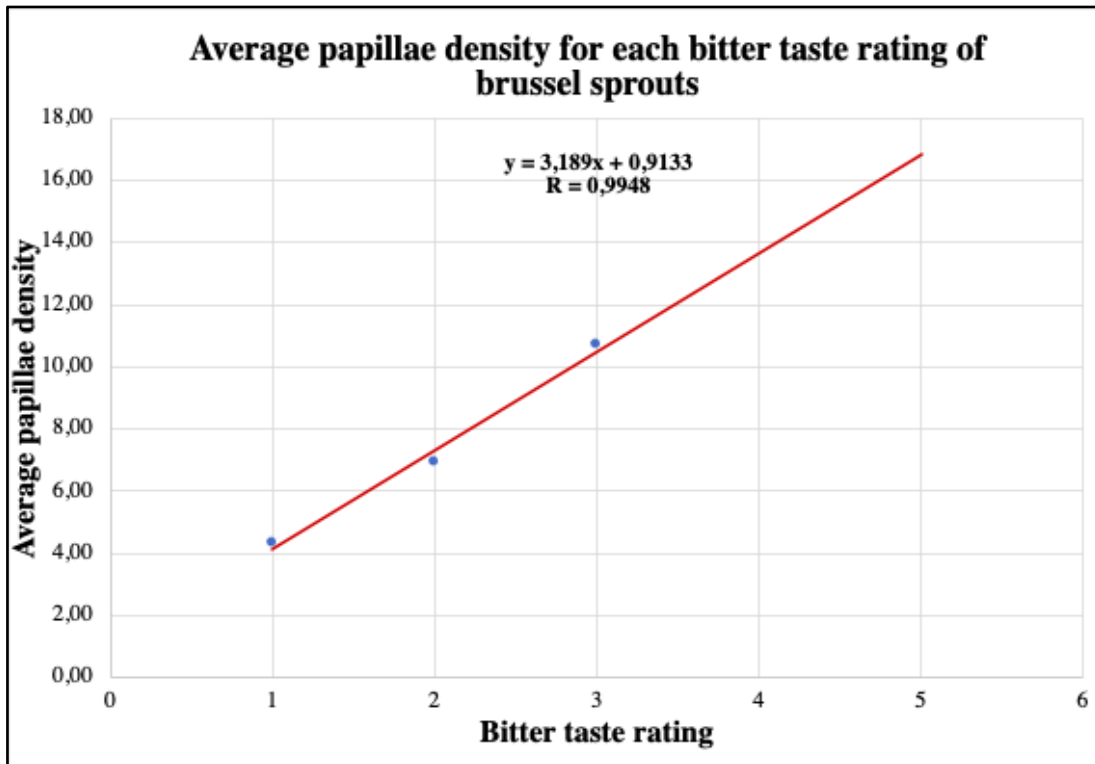
2. Broccoli

Figure III. Linear graph displaying the relationship between bitter taste rating and average tongue papillae density for broccoli



3. Brussel sprout

Figure IV. Linear graph displaying the relationship between bitter taste rating and average tongue papillae density for Brussel sprouts

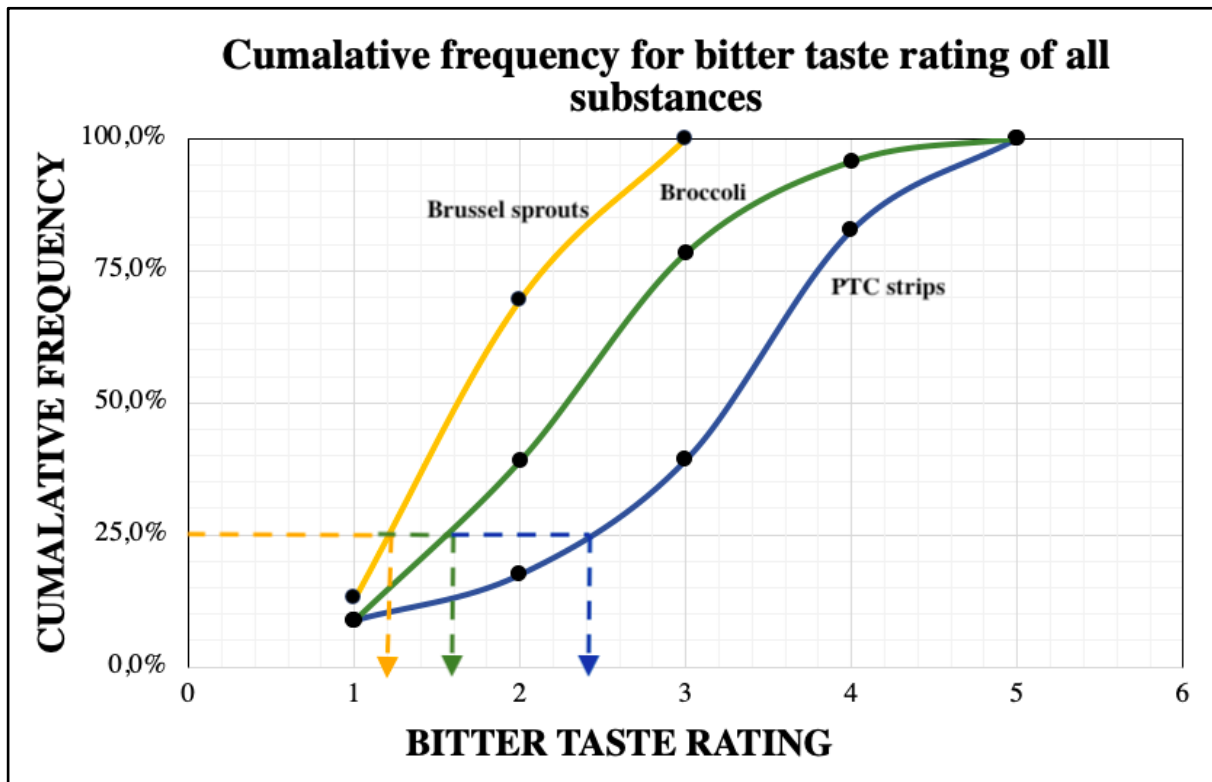


Discussion/analysis

Referring to all the plotted linear graphs above displaying the relationship between fungiform papillae density and bitter taste rating for each substance, we can conclude that there is a strong correlation between bitter taste rating and fungiform papillae density. All calculated R-values are close to 1, indicating a good fit. Thus, supporting our hypothesis that higher fungi papillae density leads to higher bitter taste sensitivity. Further, the calculated linear functions for both PTC strips ($y = 2.029x + 0.901$) and broccoli ($y = 2.1158x + 2.297$) suggests that there is approximately a 2 cm^2 increase in papillae density related to one level higher in the bitter taste rating. However, the linear function for brussel sprouts ($y = 3.819x + 0.9113$) suggests that the bitter taste rating of 1 relates to approximately 4 cm^2 (3.8 as seen in the calculated linear function).

Demonstrating classic Mendelian manner

Figure V. Cumulative frequency graph displaying the bitter taste rating of all chemicals tasted



Discussion/analysis

If the results follow a classic Mendelian ratio, 3:1 there should be 25% non-tasters within the population. For Brussel sprouts we conclude that the 25% lowest bitterness rating was lower than 1,2. For broccoli the 25% lowest bitterness rating was below 1,5 and for the PTC the 25% lowest bitterness rating was as high as 2,5. This is indicating that if my population were to follow a 3:1 ratio, the non-tasters have rated the bitterness to be higher than 1.

Statistical test: Chi-Square test to confirm Mendel's law

A chi-square test was conducted on every tested variable (broccoli, Brussel sprouts and PTC) to confirm Mendel's law checking whether the results follow a classic monohybrid inheritance with a ratio of 3:1.

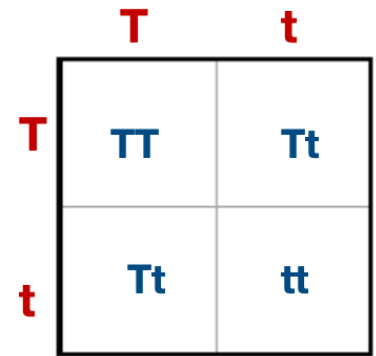
Possible alleles assuming it follows a classic monohybrid

inheritance:

TT (homozygous dominant)

Tt (heterozygous)

tt (homozygous recessive)



25% - Homozygous dominant (super taster)
 25% - Homozygous recessive (non-taster)
 50% - Heterozygous (moderate tasters)

The results of the Punnett square shows a ratio of:

3:1 where 75% = tasters and 25% = non-tasters

Null hypothesis: It is expected that the results will follow a 3:1 ratio

Alternative hypothesis: Bitter tasting perception does not follow monohybrid cross 3:1 ratio.

In these tests we have assumed that non-tasters rated 1, thus:

KEY:

Rating 1 = non-taster

Ratings 2-5 = tasters

1. Chi-square test for PTC strips and Broccoli (same results)

PTC strips					
Phenotype	Observed	Expected (E)	(O-E)	(O-E) ²	(O-E) ² / E
Taster	21	17.25	3.75	14.06	0.82
Non-taster	2	5.75	-3.75	3.06	2.44
				$\chi^2 = 3.26$	

Observations:

$$D.f. = 2-1 = 1$$

Our calculated p-value is $P(\chi^2 \geq 3.26) = 7\%$ (calculated on Excel)

From the table the critical value is 3.84 for 5% probability.

Our χ^2 is lower than the critical value (calculated to be 7%), therefore we cannot reject the null hypothesis. While the chi-square value is close the null hypothesis, we cannot be rejected at 5% probability, or confidence level.

2. Chi-square test for Brussel sprouts

Brussel sprouts					
Phenotype	Observed	Expected (E)	(O-E)	(O-E) ²	(O-E) ² / E
Taster	20	17.25	2.75	7.56	0.44
Non-taster	3	5.75	-2.75	7.56	1.32
				$\chi^2 = 1.76$	

Observations:

$$D.f. = 2-1 = 1$$

Our calculated p-value is $P(\chi^2 \geq 1.75) = 18.5\%$ (calculated on Excel)

From the table the critical value is 3.84 for 5% probability.

Our χ^2 is lower than the critical value (calculated to be 18.5%), therefore we cannot reject the null hypothesis.

Hardy-Weinberg principle

The Hardy-Weinberg method was used to determine whether the gene frequencies of the tested population were in equilibrium. The Hardy-Weinberg principle states that genetic variation in a population will remain constant from one generation to the next in absence of disturbing factors such as: mutations, natural selection, non-random mating, genetic drift and gene flow (Hardy-Weinberg equilibrium | Learn science at Scitable, 2014). We should assume that the gene for bitter tasting ability is not evolving and that the allele frequency stays constant across generations.

The Hardy-Weinberg principle states that if there are two alleles in a population then:

$$p^2 + 2pq + q^2 = 1$$

***p is the homozygous dominant genotype; q is the homozygous recessive genotype and**

2pq the frequency of the heterozygous genotype

I am assuming that all the non-tasters are homozygous recessive non-tasters (**tt**). With this basis I can calculate the percentage homozygous dominant (**TT**) supertasters and heterozygous (**Tt**) moderate tasters.

Calculations: Assuming the homozygous recessive non-taster allele frequency only

Assuming that the non-tasters rated 1, 8% of my sample population from the PTC and broccoli cases, are homozygous recessive non-tasters. Calculations can be made to determine the percentage of heterozygous moderate tasters and homozygous dominant supertasters within the population.

$$tt = 8\% \text{ or } 0.08$$

$$t = \sqrt{0.08} = 0.28$$

Therefore:

$$T = 0.72 \text{ (knowing that } T + t = 1)$$

Genotype	Genotypic frequency
TT	$0.72 \times 0.72 = 0.52$
Tt	$0.28 \times 0.72 \times 2 = 0.40$
tt	0.08

Assuming alleles based on indicated bitter taste ratings:

KEY:

TT = Homozygous dominant super taster allele (4,5)

Tt = Heterozygotes moderate tasting allele (2,3)

tt = Homozygous Recessive non-taster allele (1)

1. PTC Strips

Calculations: Assuming that the dominant TT allele has a rating of 4 and 5, I

calculated:

P = frequency of TT + ½ of frequency of Tt

$$P = 0.61 + 1/2 \times 0.30 = 0.915$$

q = 1 - P

$$q = 1 - 0.915 = 0.085$$

$$p^2 = 0.915^2 = 0.837$$

$$q^2 = 0.085^2 = 0.007$$

$$2pq = 2 \times (0.085 \times 0.915) = 0.155$$

Genotype	Genotype frequency
TT	$14 \div 23 = 0.61$
Tt	$7 \div 23 = 0.30$
tt	$2 \div 23 = 0.08$

$$p^2 + 2pq + q^2 = 0.837 + 0.155 + 0.085 = 1.07$$

Observations:

Based on my assumptions, I cannot conclude that the ability to taste PTC strips follow the Hardy-Weinberg principle with a calculated result of **1.07**. The calculations for the genotypic frequencies seen in the table above assumes that the majority of the population are supertasters (**TT homozygous dominant**). However, we can conclude that some of the individuals who rated the bitterness at 4 or 5 might actually have been heterozygous (**Tt**).

2. Broccoli

P = frequency of TT + ½ of frequency of Tt

$$P = 0.22 + 1/2 \times 0.70 = 0.57$$

$$q = 1 - P$$

$$q = 1 - 0.57 = 0.43$$

$$p^2 = 0.57^2 = 0.325$$

$$q^2 = 0.43^2 = 0.185$$

$$2pq = 2 \times (0.57 \times 0.43) = 0.490$$

Genotype	Genotype frequency
TT	$5 \div 23 = 0.22$
Tt	$16 \div 23 = 0.70$
tt	$2 \div 23 = 0.08$

$$p^2 + 2pq + q^2 = 0.325 + 0.490 + 0.185 = 1.000$$

Observations:

Similarly, we cannot conclude that the bitter tasting sensation of broccoli follows the Hardy-Weinberg Equilibrium principle with a calculated result of **1.25**. Assuming the homozygous dominant supertasters (TT) rated 4 and 5, 0.22 were supertasters, and 0.70 were **heterozygous moderate tasters (Tt)**. Based on my results, many of the individuals within the population who have given a bitter rating between 2,3 might actually have been supertasters.

3. Brussel sprouts

P = frequency of TT + ½ of frequency of Tt

$$P = 0 + 1/2 \times 20 = 10$$

$$q = 1 - P$$

$$q = 1 - 10 = -9$$

$$p^2 = 10^2 = 100$$

$$q^2 = -9^2 = 81$$

$$2pq = 2 \times (10 \times -9) = -180$$

Genotype	Genotype frequency
TT	$0 \div 23 = 0$
Tt	$20 \div 23 = 0.86$
tt	$3 \div 23 = 0.131$

$$p^2 + 2pq + q^2 = 100 + -180 + 81 = 1$$

Observations:

Based on the calculations above, we can conclude that the bitter tasting sensation for Brussel sprouts follows the Hardy-Weinberg equilibrium principle with a calculated result of 1.

However, looking at the genotypic frequencies calculated, no one claimed to be supertasters for Brussel sprouts, with no rating above 3. Consequently, these results may be inaccurate.

Considering the generally lower rating for the Brussel sprouts, this could indicate that the results were unreliable. It should also be noted that the Brussel sprouts were frozen, which could have impacted the bitterness rating.

A Hardy-Weinberg test was also conducted on the bitter taste ratings of PTC from the family population combined with the original population which had tested bitterness of PTC, giving us a larger test population.

Calculations:

P = frequency of TT + ½ of frequency of Tt

$$P = 0.51 + 1/2 \times 0.37 = 0.695$$

q = 1 - P

$$q = 1 - 0.695 = 0.305$$

$$p^2 = 0.695^2 = 0.483$$

$$q^2 = 0.305^2 = 0.093$$

$$2pq = 2 \times (0.305 \times 0.695) = 0.424$$

Genotype	Genotype frequency
TT	26 ÷ 51 = 0.51
Tt	19 ÷ 51 = 0.37
tt	6 ÷ 51 = 0.11

$$p^2 + 2pq + q^2 = 0.483 + 0.424 + 0.093 = 1.00$$

Observations:

Referring to the calculations, we can conclude that as a whole population the ability to taste PTC strips, follows the Hardy-Weinberg equilibrium principle with a calculated result of exactly 1. This means that the phenotype for bitter taste perception of PTC strips will be the same from one generation to another.

Statistical test: Investigating Hardy-Weinberg within families

A chi-square test was also conducted on the 7 families to determine whether there is a difference among the phenotypic frequencies between the parent and the children generation.

Null hypothesis: The offspring will have the same phenotypic frequency as their parents.

Alternative hypothesis: The offspring will have a different phenotypic frequency compared to their parents

In this test we also included the moderate tasters, and have assumed:

KEY:

1 - Non-taster

2-3 - Moderate taster

4-5 - Supertaster

Expected for child					
Phenotype	Observed	Expected (E)	(O-E)	(O-E) ²	(O-E) ² / E
Supertaster	8	4	4	16	4
Moderate taster	5	7	-2	4	0.57
Non-taster	1	3	-2	4	1.33
				$\chi^2 = 1.90$	

Observations:

$$D.f. = 3 - 1 = 2$$

Our calculated p-value is $P(\chi^2 \geq 1.90) = 38.6\%$ (calculated on excel)

This means that there is approximately a 38.6% chance of getting my collected results.

From the table the critical value is 5.99 for 5% probability. Therefore, we cannot reject the null hypothesis.

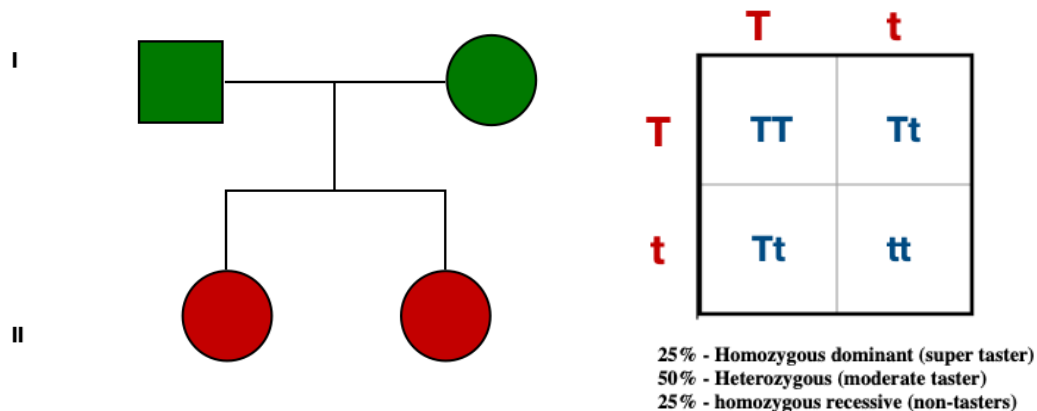
Pedigree charts: Bitter taste inheritance among families

Pedigree charts were constructed displaying the inheritance of bitter taste perception in PTC among families studied for my investigation.

KEY:
1 - Non-taster (Blue)
2-3 - Moderate taster (Green)
4-5 - Super taster (Red)
Square - male Circle – female

1. Family 1

	Mom	Dad	Daughter 1	Daughter 2
Bitterness of PTC	3	3	4	5

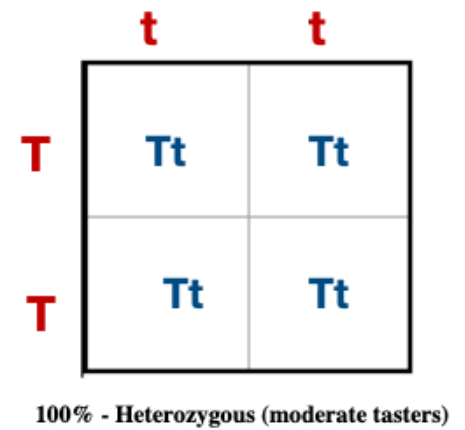
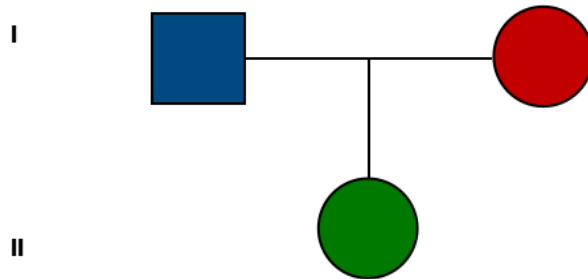


Observations:

Both parents are moderate tasters. Assuming that they are both carriers of the **heterozygous moderate tasting allele (Tt)**, there is a 75% chance that all offspring will have the ability to taste bitterness in PTC; with a 25% chance of the offspring being moderate tasters or supertasters. Referring to the pedigree chart, we see that both daughters are supertasters. This is only possible if both parents were carriers were carriers of at least one dominant tasing allele.

2. Family 2

	Mom	Dad	Daughter 1
Bitterness of PTC	5	1	3



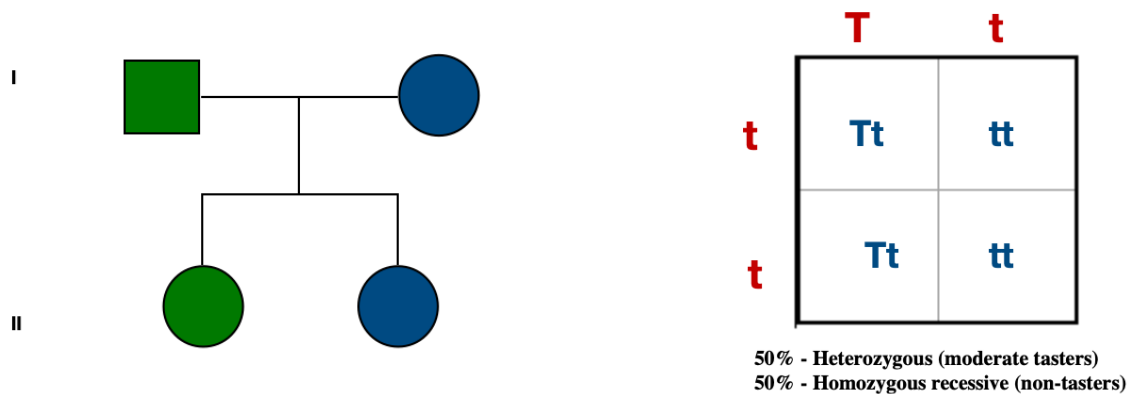
Observations:

The father is a non-taster (**tt**) and the mother is a super taster (**TT**). Assuming the mother is a carrier of the **homozygous dominant tasting allele (TT)** there is a 100% chance that all offspring produced within this family will have the ability to taste bitterness in PTC. There is no way of producing a non-taster within this family, the super tasting allele will be expressed while the effect of the other recessive allele will be masked.

3. Family 3

Mom	Dad	Daughter 1	Daughter 2
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Bitterness of PTC	1	2	2	1
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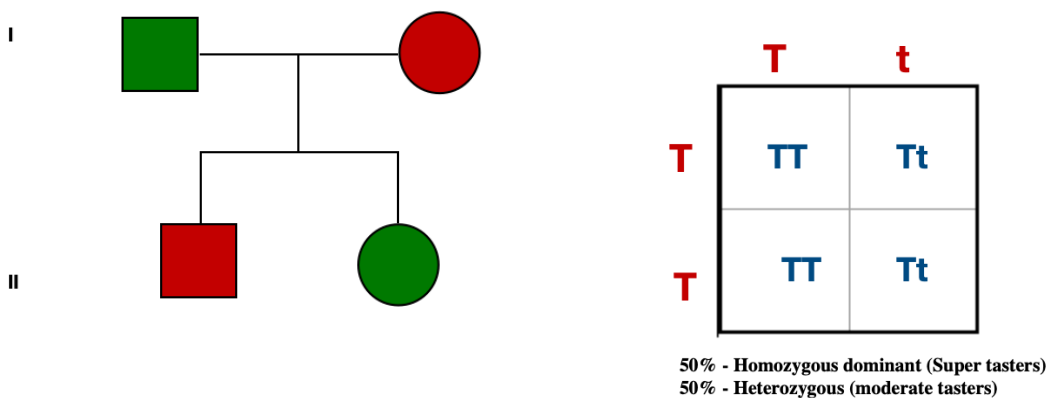


Observations:

The mother is a non-taster gene (**tt**) while the father has the ability to taste bitterness in PTC (**Tt**). One of their daughters is a taster while the other is a non-taster. The only way two parents could produce an offspring unable to taste PTC is for the dad to carry one of the taser alleles (**Tt**). The father must therefore be heterozygous. Any offspring within this family would have equal chance (50%) of being able to or unable to taste PTC.

4. Family 4

	Mom	Dad	Daughter 1	Son 1
Bitterness of PTC	4	3	3	4

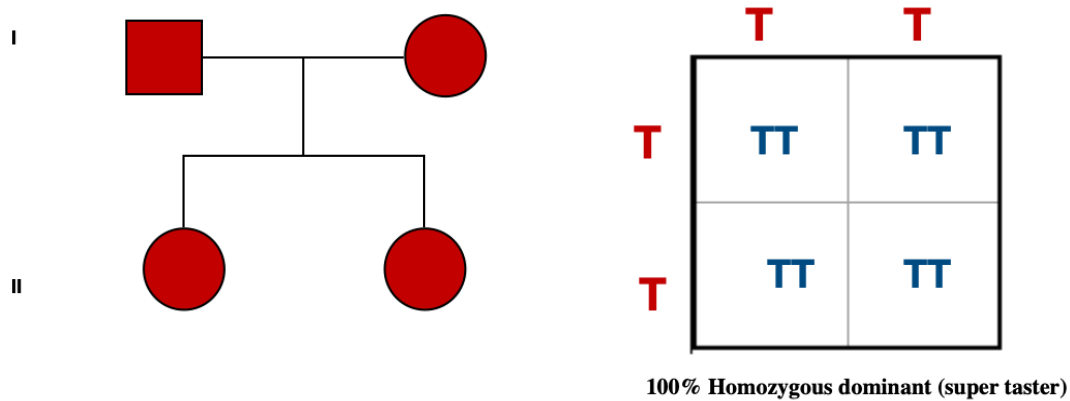


Observations:

In this family the father is a moderate taster and the mother a supertaster. Assuming that the mother is a carrier of the **homozygous dominant tasting allele (TT)** and the father a carrier of the **heterozygous moderate tasting allele (Tt)**, there is a 100% chance that the offspring will be tasters of PTC. As one of their daughters is a moderate taster, she must have inherited the recessive **t** allele from her father. Since the son is **heterozygous dominant (Tt)** he must have inherited two copies of the dominant alleles from both parents.

5. Family 5

	Mom	Dad	Daughter 1	Daughter 2
Bitterness of PTC	5	4	4	4

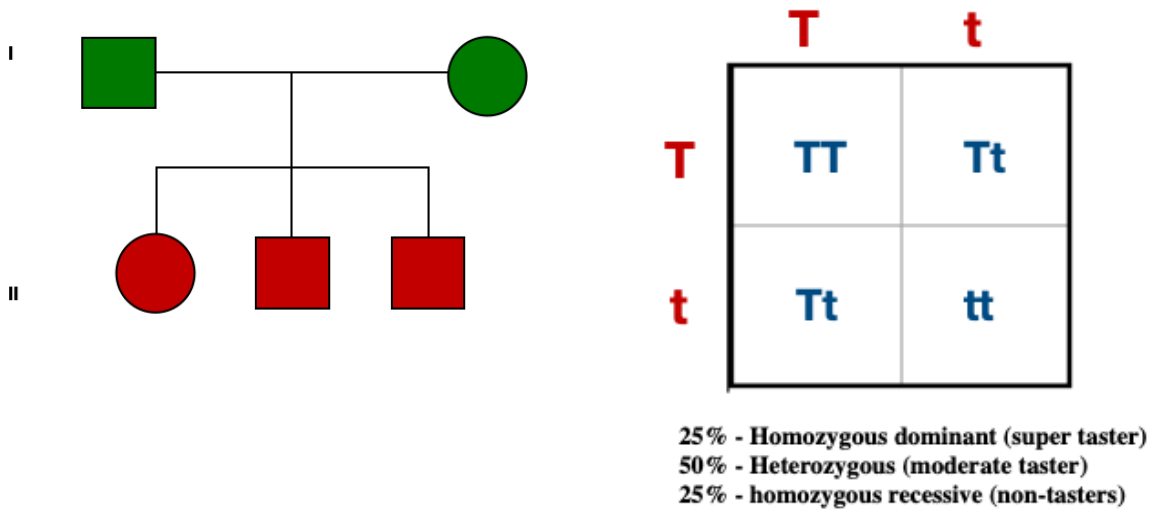


Observations:

In this family both parents are **homozygous dominant (TT)** for the bitter tasting allele. This means that any offspring produced within this family will have a 100% chance of being supertasters. There is no way of producing offspring without the ability to taste PTC strips.

6. Family 6

	Mom	Dad	Daughter 1	Son 1	Son 2
Bitterness of PTC	3	2	5	4	4

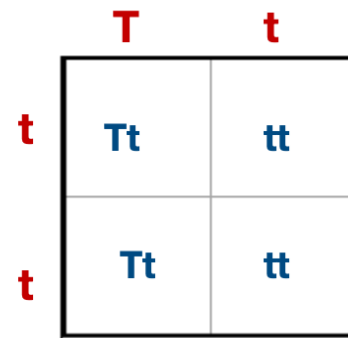
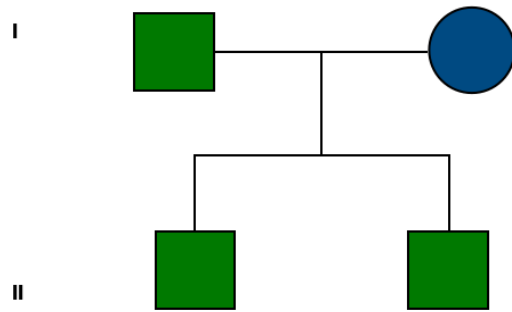


Observations:

In this family both parents are **heterozygous (Tt)** for the bitter tasting allele. This means that any offspring produced will have a 25% chance of being a carrier of the non-taster allele, 25% chance of being **dominant homozygous (TT)** supertasters and 50% chance of being **heterozygous (Tt)** for the tasting allele. As we can see in this family all offspring produced are supertasters. This means that they have all inherited a dominant tasting allele **T** from each parent.

7. Family 7

	Mom	Dad	Son 1	Son 2
Bitterness of PTC	1	2	3	3



50% Heterozygous (moderate tasters)
50% Homozygous recessive (non-tasters)

Observations:

The father is **heterozygous (Tt)** and the mother is heterozygous recessive (**tt**). Any offspring produced will have a 50% chance of being able to taste PTC strips being **heterozygous (Tt)** and a 50% chance of not being able to taste PTC **homozygous recessive (tt)**. All children within this family are moderate tasters and carriers of the **heterozygous tasting allele**. This means each child has inherited a dominant allele from their father and a recessive allele from their mother.

Conclusion

The results collected are generally in line with my proposed hypothesis. I was able to conclude that fungiform papillae density plays a significant role in bitter taste sensitivity among individuals. I found a strong correlation between bitter taste sensitivity rating and fungiform papillae density. My results are also supported by my literature reviews. An experiment closely related to mine, is a study conducted by Hayes and Bratoshuk and Duffy

(Duffy et. al 2008). They also examined the connection between bitter taste sensitivity and fungiform papillae on individuals' tongues. Individuals oral sensory phenotype was assessed by counting the number of fungiform papillae in a circular area of 6-mm in diameter on the anterior blue stained tongue surface. By using a scale participant reported bitter taste intensity of different PROP-like substances. They were concluded that the number of fungiform papillae strongly correlated with an individual's bitter taste intensity in homozygotes but not heterozygotes (Duffy et. al 2008). The findings further showed that 24% of the 198 subjects were non-tasters, 54% tasters and 22% supertasters. This study along with several others shows how bitter taste perception among individuals could depend on more than just the TASR38 gene.

Further, chi-square tests were conducted allowing me to check my hypothesis that bitter tasting ability follows a classic Mendelian ratio of 3:1. While my test sample did not replicate the 25%, 50% and 25% expected ratio of supertaster, moderate tasters and non-tasters (refer to Figure V), none of the results for the substances tested allowed me to reject the Mendelian hypothesis. I was further able to support my findings by conducting a similar experiment on families, giving me a larger population sample. As seen in all pedigree charts and Punnett grids above, all families follow a 3:1 ratio where bitter taste sensitivity is inherited in a classic Mendelian manner.

A Hardy-Weinberg statistical test was also conducted, within my population an estimated 8% of the population compared to 25% of the population considering the classic Mendelian ratio of 3:1 were non-tasters (**tt**). Considering my estimated number of non-tasters, I was able to determine whether my test population was following the Hardy-Weinberg equilibrium. The results indicated that most substances were not in equilibrium. While many were close to 1, I

was not able to accept that the population sample was following the Hardy-Weinberg principle. However, the results collected from Brussel sprouts seemed to be in line with the Hardy-Weinberg principle with a calculated value of 1. There were however several limitations regarding the Brussel sprout test, as many individuals seemed to rate very low bitter taste intensity for this substance. It is therefore difficult to make a valid conclusion with respect to the Hardy-Weinberg principle. Additionally, the Hardy-Weinberg principle was tested on a larger population, combining the PTC bitterness rating tested within the families and the rest of my test population. The results gathered allowed me to conclude that in this larger test group (population) the ability to taste PTC follows the Hardy-Weinberg principle. With a calculated result of 1, indicating that the phenotype for tasting PTC strip is stable and will not change throughout generations. This conclusion was further supported by an additional Chi-square test confirming consistency in bitterness ratings between the parents and the children generation.

Evaluation of procedure

There were several limitations to my experiment that could have been modified in order to provide more optimal results. Firstly, I was limited to the sample size, my experiment was conducted on a rather small population providing me with limited data. If there were more participants in my tests it would provide me with further analysis and more optimal results. This was clearly demonstrated when I combined the family and original test population for PTC bitterness, resulting in more optimal and consistent results.

Another challenge was to define the representative number of **homozygous recessive (tt)** tasters. Initially, I tried to determine the 25% of the population which had rated the lowest on bitterness, referring to the cumulative frequency graph in Figure V. We found that the

bitterness taste ratings varied significantly for the different substances tasted. A better assumption was therefore to label only those who had rated a bitterness of 1 as the non-tasters. Thus, in my sample the number of non-tasters was significantly lower than the predicted 25%. This could also be linked to the small sample size in my test.

Furthermore, there were several limitations to my experimental method. Due to the first experiment requiring individuals to taste 3 different substances, it is unknown whether they had any interfering flavors in their mouth or whether they had eaten before the experiment though they were instructed not to, this could have influenced their bitter taste rating; additionally there may have been some confirmation bias in the ratings given as individuals may have influenced each other sharing opinions on the bitter taste intensities. In order to allow for more optimal results, some variables should have been controlled better. Though the experiment was conducted by all participants themselves they were still conducted within small groups of people. In order to avoid confirmation biases the experiment could have been conducted individually without any other volunteers present. Furthermore, as seen throughout my analysis of results it is evident that the bitter taste rating collected for the Brussel sprouts were all relatively low. This could have been due to the fact that the Brussel sprouts were frozen and not fresh vegetables, they also seemed to be sweetened. This was another limitation to my results, as I was not able to purchase them fresh. Therefore, the data collected for Brussel sprouts were not optimal results making it difficult to draw conclusions.

Furthermore, as seen in the statistical Hardy-Weinberg test conducted, it was evident that grouping individuals into allelic groups based on their indicated bitter taste ratings is a very subjective method of gathering data. It is difficult to assume that the results are accurate and representative of each individual's alleles, an individual could be a carrier of the **dominant**

homozygous super tasting allele (TT) yet rate the bitterness much lower, such as the Brussel sprouts. Lastly, the participants' health was not taken into consideration for the experiment. Health factors and lifestyle may have affected the bitter taste intensity of each individual. Furthermore, taste and senses are said to change throughout age, this could have impacted my data collected especially when testing families. An individual could be a smoker which also contributes to bitter taste sensitivity (Da Ré et al., 2020).

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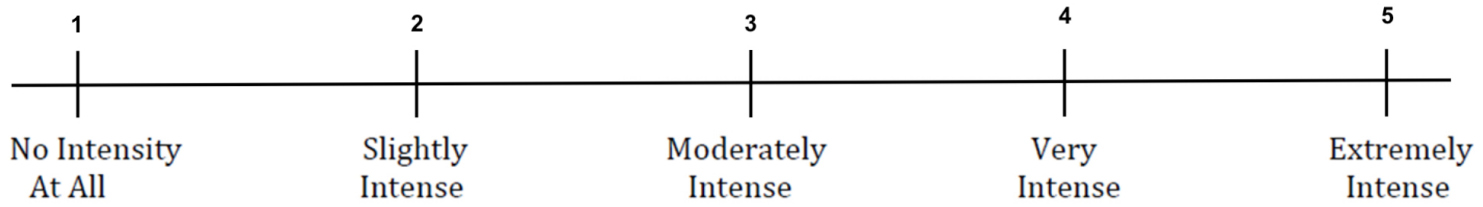
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APPENDICES

Appendix I: Bitter taste rating scale



Appendix II: Consent form volunteers

IB Biology Extended Essay Participant Consent form

Study Title: Bitter taste perception and papillae taste buds on tongue

Experimenter: XX

Description of Experiment:

In this study you will be asked to taste phenylthiocarbamide (PTC) strips as well as a series of vegetables from the Brassica Genus family rating the bitterness on an indicated scale. You will later be instructed to color your tongue blue with food coloring to allow for counting of papillae taste receptors found on your tongue.

In order to participate in this research study, it is necessary that you give your informed consent. By signing this informed consent statement you are indicating that you understand the nature of the research study and your role in that research and that you agree to participate in the research. Please consider the following points before signing:

- I understand that I am voluntarily participating in biological research;
- I understand that my identity will not be linked with my data, and that all information I provide will remain confidential;
- I understand that I will be provided with an explanation of the research in which I participated.
- I understand that certain facts about the study might be withheld from me, and the researcher might not, initially, tell me the true or full purpose of the study. However, the complete facts and true purpose of the study will be revealed to me at the completion of the study session.
- I understand that participation in research is not required, is voluntary, and that I may refuse to participate at any time further without penalty.

By signing this form I am stating that I am 16 years of age or older, and that I understand the above information and consent to participate in this study being conducted at St. Olav VGS. If I am not 16 years of age or older I must have a parent signature in order to participate.

Participant Signature: _____ **Date:** _____

Appendix III: Consent forms for families

IB Biology Extended Essay Participant Consent form

Study Title: Bitter taste perception and mode of inheritance within families

Experimenter: xx

Description of Experiment:

In this study you will be asked to taste phenylthiocarbamide (PTC) strips and rate the bitterness on an indicated scale. The results of this study will be used for my extended essay research paper which will help me analyse the mode of inheritance of bitter taste perception among individuals in a family.

In order to participate in this research study, it is necessary that you give your informed consent. By signing this informed consent statement you are indicating that you understand the nature of the research study and your role in that research and that you agree to participate in the research. Please consider the following points before signing:

- I understand that I am voluntarily participating in biological research;
- I understand that my identity will not be linked with my data, and that all information I provide will remain confidential
- I understand that I will be provided with an explanation of the research in which I participated.
- I understand that certain facts about the study might be withheld from me, and the researcher might not, initially, tell me the true or full purpose of the study. However, the complete facts and true purpose of the study will be revealed to me at the completion of the study session.
- I understand that participation in research is not required, is voluntary, and that I may refuse to participate at any time further without penalty.

By signing this form I am stating that I am 16 years of age or older, and that I understand the above information and consent to participate in this study. If I am not 16 years of age or older I must have a parent signature in order to participate.

Participant Signature: _____ **Date:** _____

Parent Signature: _____