COURSE GUIDE

BIO 201 GENETICS I (LABORATORY MANUAL)

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BIO 201

SAFETY IN THE LABORATORIES

Notes on safety in the laboratories is provided to ensure that you are aware of possible dangers in the laboratory and to inform you of the means provided to prevent accident occurring

- 1. Provision of first aid box.
- 2. Display of posters to indicate the possible dangers of chemicals and the correct remedial action to be taken in case of an accident.
- 3. Provision of fire extinguishers and blankets.
- 4. All accidents should be reported to the laboratory technologists for necessary actions to be taken.
- 5. Smoke and eating are **not** allowed in the laboratory in the interest of safety.
- 6. Fire alarm: In case of fire, the alarm will ring continuously and the building must be totally evacuated. The assembly point for this.
- 7. No horse-play (rough, boisterous fun) is allowed in the laboratory.
- 8. If you are in any doubt about the safety of a chemical, or a piece of apparatus, do not hesitate to ask a member of staff before using it.

INSTRUMENTS REQUIRED FOR ALL PRACTICAL CLASSES

- Sharp HB pencil
- Scalpel or Razor blade
- Hand lens/Magnifying glass
- A pair of forceps
- Ruler/Eraser
- Lab coat

GUIDELINES FOR PRACTICAL CLASSES

- 1. All drawings should be large to avoid obscuring important details.
- 2. No great artistic ability is needed, but the majority requirements are **accuracy** and **clarity**.
- 3. All pencils should be properly sharpened before and during practical work.
 - Possession of two or more sharp pencils will save valuable time.
- 4. The lines should be thin and clearly visible. They should not be wooly.

- 5. Drawings should not be shaded.
- 6. Always label drawings clearly and fully, especially when you are instructed to make labeled drawings.
- 7. Wherever possible, the guidelines leading from parts of the drawing to the labels should be horizontal and parallel to the top edge of the paper. The lines must never cross each other and should be ruled using a ruler.
- 8. The labels may be placed on either side of the drawing so that the lines are kept short and precise.
- 9. All drawings must bear a heading stating what the diagram is.
- 10. All drawings must bear a suitable scale or magnification or reduction below eg. x1, x5, x10, x1/2, x1/4.
- 11. Drawings should be proportionate to the object.

BASIC RULES FOR FIELDWORK

- ✓ Understand the objective of the fieldwork, the potential hazards and appropriate response to such hazards.
- ✓ Your work must be designed carefully. Do not overestimate what can be achieved because fieldwork is more tasking than laboratory work.
- ✓ Conduct a feasibility or reconnaissance or pre-field trip visit. This is done by the lecturer/field trip coordinator.
- ✓ Any physically challenged student should report to the coordinator/ organisers for appropriate precaution.
- \checkmark Never work alone without permission of the lecturer in charge
- \checkmark Make sure you can read a map and compass
- ✓ Shoes, clothing and equipment must be suitable for weather and condition you are likely to encounter.
- ✓ Avoid the use of perfumes, deodorants and scents/ scented creams and grease. These chemicals may attract bees, wasp and other insects to "pollinate you"!
- \checkmark Check weather forecast before field trip
- ✓ Details of working location, routes and time must be made known and never change arrangement

✓ International distress signals include:

E.g. six long whistle blast

Torch flashes, arm waving or shouts for help, then repeat several times Fire red flares or smoke.

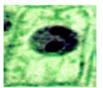
✓ Medically unstable individuals (Pregnant students, asthmatic, diabetic, claustrophobic, hydrophobic etc.) must be reported to the field trip coordinator.

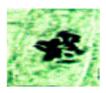
LAB I MITOSIS

Introduction

Cells arise only from division of preexisting cells. Cell division (cytokinesis) is preceded by division of the nucleus (karyokinesis). There are two forms of nuclear divisions; mitosis and meiosis. In the present Lab, we will focus on mitosis. Multicellular organisms are able to grow, repair and replace damaged tissues through mitosis. Mitosis is a process during which cells replicate their genetic materials and then divide such that one cell gives rise to two genetically identical daughter cells, each containing identical set of chromosomes to the parent cell. This adds new cells during development and replaces old and worn-out cells throughout your life. It is also a means of asexual reproduction in some organisms.

Mitosis and meiosis are preceded by interphase during which DNA and organelles are duplicated and energy is stored as ATP for use during nuclear division. Interphase is often included in discussions of mitosis, but interphase is technically not part of mitosis, but rather encompasses stages G1, S, and G2 of the cell cycle. The events occurring within the nucleus are usually observed in cells which have been fixed and stained, thus providing a series of 'snapshosts' of the stages involved in mitotic cell division. Some stages of mitosis in a dividing cell are shown below.





Interphase (dark mass)



Prophase (Chromosomes visible but not recognized)





Metaphase (chromosomes lining up along equators) Anaphase (pulling apart)

Telophase (two nuclei) **Figure 1:** Stages of mitosis

Materials

- i. Micrographs/models of different stages of mitosis
- ii. Slides of different mitotic stages
- iii. Microscope
- (A) Examination of Micrographs/ Models

Procedure

• Examine some micrographs/models of different stages of mitosis provided.

(B) Micrographic Examination of Slides

Procedure

- You are provided with slides of different mitotic stages
- Mount the slides unto a microscope and examine using low and high power objectives.

Questions:

- Identify each mitotic stage on the micrographs provided and give reasons for your answer (4 marks)
- Draw under high power objective and identify each mitotic stage on the slides provided, giving reasons for your answer. (8 marks; 2 per stage)
- 3. If a plant of genotype Aa;Bb;Cc;Dd is selfed and the genes assort independently, how many different genotypes will be found among the progeny? Show your working. (**3 marks**)

ANSWERS

(A) Examination of Micrographs/ Models

1. Identification of the mitosic stages on the micrographs provided

| S/N | Micrographs/ Models | Stage of Nuclear division (eg. anaphase of mitosis) |
|-----|---------------------|--|
| 1. | | |
| 2. | | |
| 3. | | |
| 4. | | |
| 5. | | |
| 6. | | |
| 7. | | |
| 8. | | |
| 9. | | |
| 10. | | |
| 11. | | |
| 12. | | |
| 11. | | |
| 12. | | |

(B) icrographic Examination of Slides

2. Drawings and identification of the mitotic slides provided with reasons

3. The different genotypes will be found among the progeny:

Coordinator's Signature: Date:

LAB II MEIOSIS

Introduction

Meiosis consists of two divisions; meiosis I and meiosis II. Meiosis consists of two separate divisions; meiosis I and meiosis II with only one DNA replication phase. The first division separates homologous chromosomes while the second division separates sister chromatids. In meiosis I homologous chromosomes are separated while sister chromatids are separated, During meiosis, a diploid parent cell divides to form four haploid daughter cells generally distinct from each other and from the parent cell. Meiosis is used for production of gametes, sex cells, or (sperms and eggs) in animals and during spore formation in plants during sexual reproduction. Its goal is to make daughter cells with exactly half as many chromosomes as the parent cell so that when fertilization occurs, the diploidy of the parents will be restored. It is otherwise known reduction division

Meiosis follows phases similar to those in mitosis. However, the cell has a more complex task. In prophase I of meiosis, the replicated homologous pair of chromosomes comes together in the process called synapsis to form bivalents. Non sister chromatids can also exchange segments in a process known as crossing over during which the combination of genetic information carried on the chromatids are exchanged such that the resultant chromosomes contain genes from both parents. This forms the basis that sexual reproduction introduces genes combinations to a species. Some stages of meiosis in *Secale cereale* Pollen Grain re shown in Fig. 2 below.

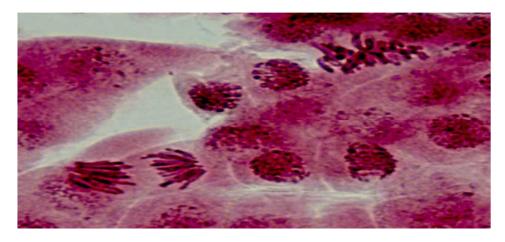


Figure 2: Meiosis of *Secale cereale* Pollen Grain showing several stages of meiosis

Materials

- i. Micrographs/models of different stages of meiosis
- ii. Slides of different meiotic stages
- iii. Microscope

(A) Examination of Micrographs/ Models

Procedore

• Examine some micrographs/models showing different stages of meiosis provided.

(B) Micrographic Examination of Slides

Procedore

- You are provided with slides of different meiotic stages
- Mount the slides unto a microscope and examine using low and high power objectives.

Questions

- Identify each meiotic stage on the micrographs provided and give reasons for your answer (5 marks)
- Draw under the high objective and identify each meiotic stage on the slides provided, giving reasons for your answer. (8 marks; 2 per stage)
- 3. Give five differences between mitosis and meiosis. (2.5 marks)
- 4. In the cross between a female AaBbccDdee and male AabbCcDdee, what proportion of the progeny will be phenotypically identical to the female parent? (Assume independent assortment of all genes and complete dominance). Show your working (**3 Marks**)
- 5. A couple in Corea are both heterozygous (Aa) for the autosomal recessive allele for albinism. They have two children, what is the probability that:
 - i. both children will be normal? (**0.5 marks**)
 - ii. both children will be albinos? (0.5 marks)

iii. both children phenotypically identical with regard to skin color? (0.5 marks)Show your working (2 marks)

ANSWER

(A) Examination of Micrographs/ Models

1. Identification of the meiotic stages on the micrographs provided

| S/N | Micrographs/ Models | Stage of Nuclear division (eg. anaphase of meiosis I) |
|-----|---------------------|--|
| 1. | | |
| 2. | | |
| 3. | | |
| 4. | | |
| 5. | | |
| 6. | | |
| 7. | | |
| 8. | | |
| 9. | | |
| 10. | | |
| 11. | | |

| 12. | |
|-----|--|
| | |
| | |

(B) Micrographic Examination of Slides

2. Drawings- and identification of the meiotic stages on the slides provided with reasons

3. Differences between mitosis and meiosis

| S/N | MITOSIS | MEIOSIS |
|-----|---------|---------|
| 1. | | |
| 2. | | |
| 3. | | |
| 4. | | |
| 5. | | |

4. Phenotypic proportion that are identical to the female parent:

5. i. The probability that both children will be normal

ii. The probability that both children will be albinos:

iii. The probability that both children phenotypically identical with regard to skin color:

Coordinator's Signature: Date:

LAB III MONOHYBRID CROSSES AND THE PUNNETT SQUARE

Introduction

Genetics is concerned with transmission and expression of genes. These are factors (genes) that control the function, development and ultimate appearance of individuals. In this Lab, you will examine Mendel's First Law of inheritance (the law of segregation) as illustrated by monohybrid cross. According to the law, the diploid (2n) condition in higher organism contains two alleles of each gene. During meiosis, these alleles segregate, hence only one of them enters each gamete (n). Fertilization restores the diploid (2n) condition in the zygote. Mendel was the first to recognize the 3:1 phenotypic ratio as a pattern of inheritance for monohydrid crosses.

A grid-like tool (Punnett Square) is used to make predictions about various genetic problems. The Punnett Square shows only the probability of what might occur and not the actual results; the chance of something occurring. The Punnett Square only shows the chances of what might occur each time the event is undertaken

In this investigation, we shall compare the expected result from a monohybrid cross with the actual result from a Mendel monohybrid cross using beans seeds of different colours to represent the alleles. A Punnett Square will be used to predict the possible genotypes and phenotypes and their ratios from a monohybrid cross.in order to assess the agreement with the expected ratios.

Materials

- ·i. Brown beans seeds
- ii. White beans seeds
- iii. Small paper bags (sone labeled male and others labeled female)

Procedure

• You are provided with small paper bags labeled male and female. Each paper bag contains 30 beans seeds; 15 white beans seeds and 15 brown bean seeds. A seed represent an allele such that brown is dominant (B) while white (b).

- Students shall form groups of three. Each group comprising of a male and a female student representing two heterozygous parents (Bb x Bb), mating pair and the third either a male or female student.
- Each group will pick up two paper bags (a male and a female).
- A male student in the group will be in charge of the male bag and a female student in charge of the female bag while the third student will serve as a recorder.
- The members of the mating pair are to shake the envelopes thoroughly to mix the seeds, before producing gametes by making blind selection of a seed each from the bags to form a zygote. The possibilities that can be made from this selection are BB, Bb or bb
- Record the resulting genotype and phenotype in a data Table. (2 marks)
- Returned the seeds back into to their respective bags and shake thoroughly before another selection.
- Each group should make a total of 20 trials.
- Record the result of your group on the white board such that members of other groups will have access to it in order to compare the class total.

Questions

1. Determine:

| i. | Phenotypic ratio | (1 mark) |
|----|------------------|----------|
|----|------------------|----------|

- ii. Genotypic ratio (1 mark)
- 2. How do this phenotypic ratio compare with the expected ratio of 3:1? (1 mark)
- 3. Show the cross of the parents given in the above procedure using a Punnett Square. (2 marks)
- 4. Determine the following for the class totals (2 marks)
 - i. Genotypic ratio
 - ii. Phenotypic ratio
- 5. How close is the phenotypic ratio to the expected 3:1 ratio when the results for your group and that of the class total are compared? Explain why this is so. (2 marks)

Proper recording of results (1mark) ANSWERS

| | Data Table | |
|-------|----------------------|-----------------------|
| Trial | Offspring's Genotype | Offspring's Phenotype |
| 1 | | |
| 2 | | |
| 3 | | |
| 4 | | |
| 5 | | |
| 6 | | |
| 7 | | |
| 8 | | |
| 9 | | |
| 10 | | |
| 11 | | |
| 12 | | |
| 13 | | |
| 14 | | |
| 15 | | |
| 16 | | |
| 17 | | |
| 18 | | |
| 19 | | |
| 20 | | |

Determination of:

1. Phenotypic ratio

ii. Genotypic ratio

2. Comparison of the phenotypic ratio compare with the expected ratio of 3:1

3. Cross of the parents given in the above procedure in a Punnett Square.

Male _____ X Female _____

Result of the class totals

| Group | BB | Bb | bb |
|-------|----|----|----|
| 1 | | | |
| 2 | | | |
| 3 | | | |
| 4 | | | |
| 5 | | | |
| 6 | | | |
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| N | | | |
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- 4. Determination of the following for the class totals:
- i. Genotypic ratio

ii. Phenotypic ratio

| 5. | Correlation between the phenotypic ratio to the expected 3:1 ratio when the results for your group and that of the class total are compared |
|-------|---|
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INTRODUCTORY DERMATOGLYPHICS LAB IV

Introduction

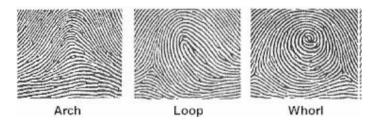
Dermatoglyphics refer to the study of ridged skin patterns (dermatoglyphs) which can be found on finger prints, palms, toes and sole of primates and some other mammals. Dermal ridges on the finger print are formed early in foetal development (13 to 19 weeks after fertilization) and remained unchanged throughout life. Although the pattern of dermal ridges is largely genetically determined but it is also subject to local environmental modifications during critical period of ridge modification.

The human hands dominate all other organs in terms of relative importance and are sources of physiological and psychological information waiting to be tapped, hence its increasing importance in the field of dermatoglyphics.

There are two general parts of print patterns:

- i. Finger print pattern
- ii. Palmer landmarks

In this investigation, we shall consider the finger print patterns. There are three major finger print patterns, namely; Arch, Loop and Whorl.



These three types have been subdivided into various subgroups such as simple arch, tented arch, ulnar loop, radial loop and spiral whorl as shown below.



simple arch tented arch

ulnar loop

spiral whorl

radial loop

For this exercise, we shall consider only the general types.

Materials

- i. Methylated spirit
- ii. Inkpad
- iii. Clean white sheet of paper

Procedure

- Clean your hand with methylated spirit to avoid greasy material which might distort the dermatoglyphic patterns.
- Press the finger to be finger printed on the inkpad.
- Roll the finger from nail to nail on a clean white sheet of paper.
- Then score the dermal count by drawing a straight line between the triradius and the core of the finger print pattern.
- For clarity magnified the pattern by viewing under a magnifying lens

Questions

- Classify the finger print pattern for each of the ten fingers.
 (5 marks)
- 2. Compare your finger print with those of two named members of your class and comment on the differences. (4 marks)
- 3. Two black mice are crossed, ten black and three white offspring result
 - i. Which allele is dominant and which is recessive? Give reasons for your answer (2 marks)
 - ii. Deduce the genotypes of the parents. (1 mark)
 - iii. How will the phenotypic ratio be modified if the alleles are codominant? (**2 marks**)

ANSWERS

1. Classification of the finger print pattern for each of the ten fingers.

| Finger | Left Hand Print | Right Hand Print |
|-----------------|-----------------|-------------------------|
| Thumb | | |
| | | |
| 2 nd | | |
| 3 rd | | |
| 4 th | | |
| 5 th | | |

2. Comparison of your finger print with those of two named members of your class and comments on the differences.

| 3. (a) The dominant allele is and the recessive is |
|--|
| |
| Reasons: |
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| |
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| |

(b) The genotypes of the parents are and

(c) If the alleles are codominant, the phenotypic ratio will be modified thus

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LAB V KARYOTYPING

Introduction

A karyotype is a photographic display of chromosomes in the cell of an organism arranged as homologous pair s according to size. It is prepared by cutting out individual chromosome photographed in a dividing cell, especially when the cell is t metaphase as the chromosomes are more distinct at this stage.

Genetic Counselors use karyotypes to identify conditions caused by extra or missing chromosomes. In this lab, you will prepare a karyotype of a human and determine the genetic characteristics of the individual.

The karyotype of a normal human male and a normal female is shown in Fig. 3.

| X | <i>K</i> | K | | 2 | | l | |
|---------------|------------------|----------------------|----|----|------------|--------------------------|--------------------------|
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| 13 | 14 | 15 | | | 16 | 17 | 18 |
| ** | | | 84 | 66 | | lſ | |
| 19 Xaryoti | 20 19e:46. | × | 21 | 22 | | X S B | ¥ |
| Male | | | | | | fema | le |

Figure 3: Karyotype of a normal human male and female

Materials

- i. chromosomes in a metaphase
- ii transparent cellotape

Procedure

- Cut the chromosomes in a metaphase spread and arrange them in homologous pairs.
- Use transparent cellotape to fix the chromosomes in the space provided.

Questions

- 1. What is the diploid chromosome number ? (1 mark)
- 2. Describe the genetic condition of the individual. (2 marks)
- 3. In a certain species of marine fishes, the caudal tail may be long or short. When certain short-tailed individuals are crossed with long tailed ones, they produce short-tailed progeny. When other sort-tailed fishes are crossed with long-tailed ones, they produce approximately equal number of long-tailed and short-tailed fishes. When long-tailed individuals are intercrossed, they always produce progenies like themselves.
 - i. What is the dominance relationship between the two alleles? (1 mark)
 - ii. Represent the above crosses with a diagram showing the genotypes and phenotypes of all individuals in the crosses (6 marks)

ANSWERS

| 1. The diploid chromosome number is | | | |
|--|---------------|--|--|
| 2 Description of the genetic condition of the individual | | | |
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| 1. The dominance relationship between the two elleles | | | |
| 4. i. The dominance relationship between the two alleles | | | |
| 4. i. The dominance relationship between the two alleles | | | |
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BIO 201

ii. Diagram showing the genotypes and phenotypes of all individuals in the crosses

Coordinator's Signature: Date:

LAB VI ABILITY TO TASTE PTC: AN AUTOSOMAL DOMINANT INHERITANCE

Introduction

PTC (Phenyl thiocarbamide), a chemical substance, has an unusual property that it either tastes very bitter or is virtually tasteless, depending on the genetic makeup of the taster. The ability to taste PTC is an autosomal dominant trait in humans. Individuals differ in their ability to taste PTC; the ability to taste is odominant over inability to taste. Tasters are thus designated as homozygous dominant (TT) or heterozygous Tt while non-tasters are homozygous recessive (tt). A very convenient way to score this phenotype is to impregnate filter paper for tasting. However, an appropriate method for advanced classes is to determine the lowest concentration that can be tasted by each individual and to plot the values on a histogram.

In this Laboratory exercise, students will determine their phenotypes and possible genotypes for PTC. They will also determine the taste thresholds for both tasters and non-tasters for the class.

Materials

- Serial dilution of PTC
- Glass rod

Procedore

- You are provided with serial dilution of PTC.
- Dip a clean glass rod into the most diluted solution and taste .
- If there is any distinctive taste, repeat the process for the next more concentrated solution and tasted.
- This should continue until the PTC is detected.

Questions

1. Determine your phenotype and your probably genotypes

(2 marks)

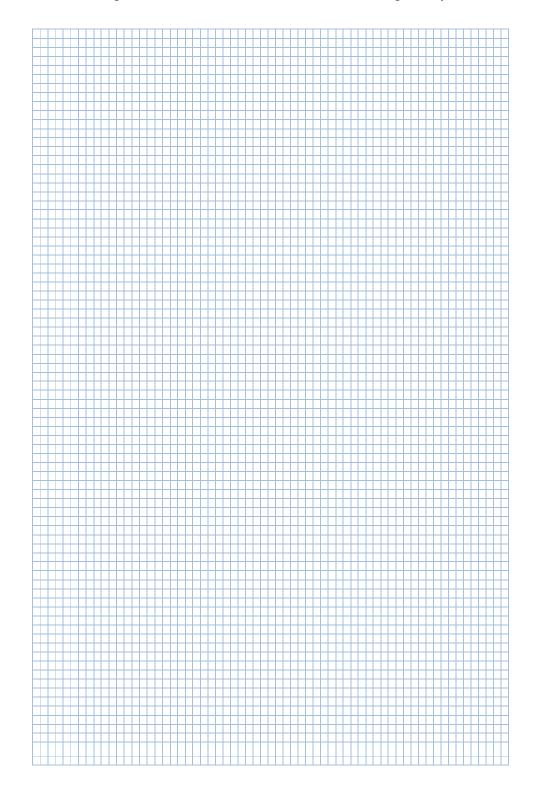
(a). Present the class distribution of PTC tasting ability in a Table (3 marks)

- (b) Use the data collected to draw a histogram of the class distribution of PTC tasting ability (4 marks). What type of distribution is presented in the graph? (1 mark)
- 3. Determine the class threshold for tasters and non-tasters. (2 marks)
- 4. In humans, differences in the ability to taste phenylthiourea are due to a pair of autosomal alleles. Inability to taste is recessive to ability to taste. A child who is a nontaster is born to a couple who can both taste the substance. What is the probability that their next child will be a taster? (1 mark). Show your working (2 marks)

ANSWERS

- 1. My phenotype is and my probably genotype (s)
- 2 (a). Table of the class distribution of PTC tasting ability

| CONCENTRATION | TALLY MARK | FREQUENCY |
|---------------|------------|-----------|
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(b) Histogram of the class distribution of PTC tasting ability

The distribution presented in the graph is

.....

- 3. Determination of the class threshold for tasters and non-tasters.
- 4. probability that their next child will be a taster?

Coordinator's Signature: Date:

LAB VII SALT TASTE RECOGNITION THRESHOLD IN HUMAN

Introduction

Salt taste sensitivity is one of the several genetic variation found in human populations. Studies on salt taste sensitivity are important in Clinical medicine and Public health because excessive salt intake has been associated with diabetes and Hypertension.

Like other taste qualities such as sweat, sour and bitter, salt taste is mediated by taste receptors found in the cell bud of the tongue. The weakest concentration at which a specific salt taste can be identified (ie. salt taste recognition threshold) varies from one person to the other.

In this Lab, the taste recognition threshold of the students in your group will be determined to obtain the salt taste sensivity distribution of the group.

Materials

i. distilled water

ii. labeled beakers

iii. filter papers

iv. fume hood

v. NaCl (salt)

Procedore

- Dissolve 3.5 g of NaCl in 100 ml of distilled water in beaker no. 1 and stire until the salt dissolves to give 3.5 percent (%) salt solution (3.5 grams per 100 ml).
- Measure-50 ml of your new 3.5% salt solution from beaker no. 1 and pour it into a new, empty beaker (no. 2).
- Add 50 ml of distilled water to this solution and stir, creating a 1.75% salt solution.
- Repeat four times to create solutions of 0.875%, 0.4375%, 0.2188% and 0.1044% salt (a total of 7 serial dillutions ranging from 0.0 ie. distilled water to 3.5).
- Soak the strips of filter paper overnight in the 7 solutions.
- Air dry the strips in a fume hood.

- Then cut the strips into smaller pieces (3cm x 2cm) and place them into labeled petri dishes until use.
- Give the filter papers to the subject starting from the lowest concentration.
- If correctly identified, give the next higher concentration until a distinct salt taste is recognized..

Questions

- 1. Record the salt taste recognition threshold of the individuals in your group. (2 marks)
- 2. Prepare the Frequency Table of the Data. (2 marks)
- 3. Plot a frequency curve for the Table (**3 marks**)
- 4. Determine your salt taste threshold value and explain the category of salt taste threshold that you belong. (3 marks)

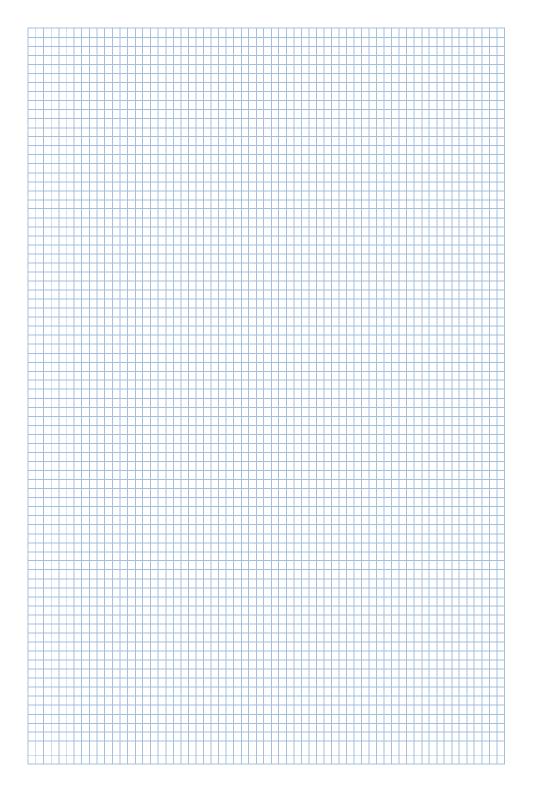
ANSWER

1. Salt taste recognition threshold of Subjects in the Group

2. The Frequency Table of the Data

| Solution | Tally Mark | Frequency |
|----------|------------|-----------|
| 1 | | |
| 2 | | |
| 3 | | |
| 4 | | |
| 5 | | |
| 6 | | |
| 7 | | |
| 8 | | |
| 9 | | |
| 10 | | |
| 11 | | |
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3. A frequency curve for the Table



4. Determination of my salt taste threshold value and explain the category of salt taste threshold that I belong.

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Coordinator's Signature: Date:

BIO 201

BIO 201