

DEPARTMENT OF CHEMISTRY

SYLLABUS

CLASS: I B.SC CHEMISTRY

Batch:2019

19CHU113

BASICS AND HYDROCARBON PRACTICAL

Semester-I
4H 2C

Instruction Hours/week:L: 0 T:0 P:4

Marks: Internal: 40 External: 60 Total:100

Scope

This course is an introduction to chemistry lab that illustrates principles of organic chemistry and laboratory techniques. The course presents the practical knowledge of the separation, purification and characterisation of organic compounds.

Objectives

The student will be able to

1. Purify organic compounds by crystallisation.
2. Characterisation of the compounds by elemental analysis, melting point, and effect of impurities on the melting point.
3. To separate organic compounds by paper chromatographic methods
4. Preparation of organic compounds.
5. The lab will also provide hands-on opportunities to develop and apply this knowledge

Methodology

Laboratory experiments, Melting point apparatus, paper chromatography, Heating mantles

1. Checking the calibration of the thermometer
2. Purification of organic compounds by crystallization using the following solvents:
a. Water, b. Alcohol, c. Alcohol-Water
3. Determination of the melting points of unknown organic compounds.
4. Effect of impurities on the melting point – mixed melting point of two unknown organic Compounds
5. Determination of boiling point of liquid compounds. (boiling point lower than and more than 100 °C by distillation)
6. Chromatography
a. Separation of a mixture of two amino acids by ascending paper chromatography

- b. Separation of a mixture of two sugars by ascending paper chromatography
- c. Separation of a mixture of o- and p-nitrophenol or o- and p-aminophenol by thin layer chromatography (TLC)

7. Detection of extra elements

8. Organic Preparations

- (i) Preparation of benzophenone oximes
- (ii) Preparation of Phenyl benzoate

Suggested Readings:

Text Books:

1. Mann, F.G. & Saunders, B.C. (2009). *Practical Organic Chemistry*. Pearson Education.

Reference Books:

1. Furniss, B.S., Hannaford, A.J., Smith, P.W.G. & Tatchell A.R. (2012). *Practical Organic Chemistry*. 5th Ed. Pearson.

DEPARTMENT OF CHEMISTRY

Title of the Paper : **BASICS AND HYDROCARBON PRACTICAL**

Paper Code : **19CHU113**

Class : **I- B. Sc-Chemistry**

Year and Semester : **2019–2020 and I-Semester**

LIST OF PRACTICALS

S.No.	Duration Hours	Name of the Experiments	Support Materials
1	4	Calibration of the thermometer	W1
2	4	Purification of organic compounds by crystallization	R1: 135-156
3	4	Determination of melting points of unknown organic compounds	R1: 236-240
4	4	Effect of impurities on melting point-mixed melting point	R1: 240-241
5	4	Determination of boiling point of liquid compounds	R2: 241-243
6	4	Organic preparations: (i) Preparation of benzophenone oximes	R1: 1040
7	4	(ii) Preparation of Phenyl benzoate	R4:416-417

8	4	Chromatography: Separation of a mixture of two amino acids by ascending paper chromatography	R3:200
9	4	Chromatography: Separation of a mixture of two sugars by ascending paper chromatography	R3:198
10	4	Detection of Extra elements	R1: 1204-1210
11	4	Viva –voce questions discussion	
12	4	Model practical examinations	

Reference Books:

1. R1: B.S. Hannaford, A.J.Smith & Tatchell.A.R, 2012, practical Organic chemistry,5th Edition, Pearson Education.
2. R2: Venkatesan, Veeraiyan Vaithianathan. B.Sc Practical Chemistry, 2002, S.Chand publications.
3. R3. Raj K Bansal, Practical Organic Chemistry 2007, S. Chand publications
4. R4: A.O.Thomas,2003, Practical chemistry for B.Sc Main Students, Scientific Book Centre, Cannanore-1, Kerala

Website:

W1.[http//www. Calibration of the thermometer](http://www.Calibration of the thermometer)

LAB MANUAL

BASICS AND HYDROCARBON PRACTICAL

(19CHU113)

I B. SC CHEMISTRY

KAHE

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Basics and hydrocarbon practical (17CHU113)

Ex.no	Title of the experiment	Page no
1	Checking the calibration of the thermometer	3
2	Determination of the melting point of the unknown organic compound	5
3	Determination Of Effects Of Impurities On Melting Point - Mixed Melting Points Of Organic Compound	8
4	Determination of boiling point of the unknown organic compound	10
5	Purification of benzoic acid by crystallization using water	12
6	Preparation of Benzophenone oxime	15
7	Preparation of Phenyl benzoate	17
8	Detection of elements present in the organic compounds	18
9	Separation of a mixture of two amino acid by ascending paper chromatography	19
10	Separation of a mixture of two sugars by ascending paper chromatography	21

Exp. No: 1 Calibration of the thermometer

Aim:

To calibrate a stem type thermometer and then use it to correct the air temperature of the laboratory.

Introduction:

One of the most common types of laboratory types of thermometer is the liquid expansion thermometer. In this type of thermometer, an expansion liquid, usually mercury or alcohol fills a glass bulb attached to a long stem with a uniformly bored expansion column. When heated the liquid expands up the bore until the bulb reaches thermal equilibrium with the bulb material whose thermometer temperature is being measured. The expansion of the liquid is such that the height it reaches in the stem is linear with temperature. This thermometer is typically marked with equal spacing along them. They are then calibrated at two different fixed point temperatures. Alcohol thermometer are lower accuracy of mercury thermometer, but are commonly used in chemistry laboratory mercury thermometer is toxic and difficult to clean up in the case of breakage. Calibration is such that a measurement is performed whose result is known. The instrument is then adjusted such that its reading gives this result. Or a correction factor is determined such that subsequent reading can be corrected by known instrument error. From stem thermometer, the markings alone the stem cannot be adjusted, a correction curve is prepared such that the temperature reading can be converted to accurate temperature known temperature bath for calibration can be generated using the ice point and the boiling point of the water. This temperature baths are called fixed points because of that use as calibration markers for thermometer.

Procedure:

Thermometer identification:

First note the temperature range of the thermometer, note whether or not temperature in thermometer is the total immersion or partial immersion type. Partial immersion thermometer will have an immersion mark and are designed so that only that part of the stem is exposed to the temperature being measured. Total immersion thermometer is designed so that both the bulb and

the entire liquid column must be exposed to the temperature being measured. Check

to make sure that the liquid in the stem of the thermometer has not separated.

Calibration at the ice point of water:

Fill the styrofoam container with crushed ice, add enough free cold distilled water to cover the ice but not so much of water such that the ice floats. Thoroughly stir the ice water mixture and hang your thermometer by stirring from a clamp attached to a ring stand until it is approximately inserted into the ice water. Allow the temperature shown by the thermometer to stabilize (approximately 10 minutes is required to establish thermal equilibrium) after 3 minutes at the stable temperature to the correct precision. The ice point of the water is remarkably stable at 0°C.

Calibration at the boiling point of water:

Setup a hot plate with the 500ml flask resting on it. The flask should be supported by a clamp from the ring stand. Fill the flask about half filled with distilled water and add a few boiling chips to promote smooth boiling. Hang the thermometer from the ring stand as before such that the immersion mark is in the neck of the flask. Turn on the hot plate and allow the water to come to its point of boiling. Allow the temperature shown by the thermometer to stabilize. After 3 minutes at the stable temperature, record the temperature to the correct precision. The boiling point of the water is extremely sensitive to the atmospheric temperature.

Air temperature:

The air temperature in the room temperature will be measured by measuring the temperature of the large water bath that has been allowed to come to thermal equilibrium with the air. Hang your stem thermometer from the ring stand into the water bath, allow for thermal equilibrium to be established. Make the appropriate reading.

Result:

Thermometer identification: temperature range =

Calibration at the ice point of water: thermometer reading =

Calibration at the boiling point of water

1. Boiling point of water =

2. Atmospheric pressure =

Air temperature =

Exp. No: 2 Determination of the melting point of the unknown organic compounds

Aim:

To determine the melting point of the given unknown organic compounds.

Introduction:

The melting point of pure solid organic compound is one of its characteristic physical properties along with molecular weight, boiling point, refractive index and density. A pure solid will melt reproducibly over a narrow range of temperature, typically less than 1 degree C. The process of determining this melting point is done on a truly microscale less than 1mg of material; the apparatus is very simple, consisting thermometer, a capillary tube to hold the sample in the heating bath or melting point apparatus.

Melting point is determined for three reasons. If the compound is known one the melting point will help to characterize the sample in hand. If the compound is known then the melting point is recorded in order to allow further characterization by others and finally the range of the melting point is indicative of the purity of the compound. An impure compound will melt over a wide range of temperatures. Recrystallization of the compounds will purify it and melting point range will decrease. In addition, the entire range will be displaced upward. For example: an impure sample might melt from 120 degree C to 124 degree C and after recrystallization melt at 125 degree C-125.5 degree C. A solid is considered pure if the melting point dose not changes after recrystallization.

A crystal is an orderly arrangement of molecules in a solid. As heated is added to a solid, the molecules will vibrate and perhaps rotate but still remain in solid .At a characteristic temperature it will suddenly acquire the necessary energy to overcome the force that attracts one molecule to another and it will undergo translational motion in other words; it will become liquid.

The forces by which one molecule is attracted to another include ionic attraction, vanderwaal's forces, hydrogen bonding, and dipole-dipole attraction. Most but by no means, organic molecules are converted in covalent in nature and melt temperature below 300 degree C. Typically inorganic compounds are ionic and have much higher melting point. eg: sodium chloride melt at 800 degree C. Ionic organic molecules often decomposed before melting, as do compounds having strong hydrogen bonds such as sucrose, other factor being equal, large molecules melt at higher temperature than small once. Among structural isomers the more symmetrical will have the higher melting point.

Experimental procedure:

Apparatus:

Melting point apparatus

The melting point apparatus consists of an electrically heated aluminum blocks that accommodates three capillaries. The sample is illuminated through the lower part and absorbed with the six power ions through the upper part. The heating range can be controlled with the thermometer. The apparatus can be used up to 360°C for above useful limited on silicon oil (about 350°C)

Sealing a melting point capillary tube:

Capillary can be obtained commercially or can be made by drawing out 12mm soft glass tubing. The tubing is rotate in the hottest part of the Bunsen burner flame until it is very soft and being to sag. It should not drawn out dving heating, but it is removed from the flam and after a moment hesitation drawn steadily and not to rapidly to arm's length with the same practice it is possible to produce 10 to 15 good tubes in a single drawing. the long capillary tube can be cut into 100mm length with a glass scorer. Each tube is sealed by rotating the end in the edge of the small flame.

Filling melting point capillary:

The dry sample ground to fine powder on a watch glass or a piece of glassine paper on a hot

surface using the flat portion a spatula. It is formed into a small pile and the melting point. Capillary forced down into the pile. The sample is shaken into the closed end of the capillary by warping sharply on the hard surface or by dropping it down a two feet length of the glass tubing onto a hard surface. The height of the sample should be no more than 2to3mm.

Determining the melting point:

The accuracy of the melting point depends on the accuracy of the thermometer so the first exercise in this experiment will be to calibrate the thermometer of the melting point of pure known compound will be determine and deviation recorded so that a correction can be applied to further melting point. We forwarded however that the thermometer are usually fairly accurate.

The most critical factor in determining an accurate melting point the temperature rise should not be greater than 1°C |minute. This may seen extraordinary slow, but it is necessary in order the heat from bath we transferred equally to the sample and to the glass and the mercury of the thermometer experience you know the rate at which the ice melts. Consider doing the melting point experiment on the ice cube. Because water melts at 0°C you would need to have a melting point bath a few degrees below zero. To observe true melting point of the ice cube you would need to raise the temperature extraordinary slowly. The ice cubes would appear to begin to melt at 0°C and if you waited for temperature equilibrium to be established it would be melted at 0.5°C If you were impatient and raise the temperature too rapidly. The ice might appear to melt over the range to 0°C to 20°C . Similarly melting point determine in capillaries will not be accurate if the rate of the heating is too fast. The rate of heating is the most important factor in obtaining accurate melting point.

As the melting point is approached the sample may shrink because of the crystal structure changes. However, the melting process begins when the first drop of the liquid is seen in the capillary and it is ends when the last trace of the solid disappears. If determinations are to be done on the two or three samples that differ in melting point by much as 10 degree C two or three capillaries can be secured to the thermometer together and the melting point is observed in

succession without removal of thermometer from the apparatus. As a precaution

against interchange of the tube, use some system of identification such as 1, 2, 3 dots made with a marking pencil. Determine the melting point of the given sample and repeat the determination and if the two determination and if the two determinations do not check within 1°C do a third one.

Result:

The melting point of the given organic compound is =

Exp. No: 3 Determinations of Effects Of Impurities On Melting Point-Mixed Melting Points Of Organic Compound

Aim:

To determine the effects of impurities on melting point mixed melting point of the given organic compound.

Introduction:

The melting point of the compound is the temperature at which the solid state is in equilibrium with the liquid phase. A solid compound changes to a liquid when the molecule acquire enough energy to overcome the forces that of holding them together in the orderly crystalline lattice for most organic compound these intermolecular forces are negatively weak. The melting point change is defined as the span of temperature from the point at which the entire first begin to liquefy to the point at which the entire sample is liquid most pure organic compounds melt over a narrow temperature range 1-2°C.

Procedure:

Make a mixtures of the given organic compounds in the approximate proportions 1:4 and 1:1 by putting side by side the correct numbers of equal sized small piles the two substance and then providing the mixture thoroughly for at least a minute on a watch glass using a metal spatula. Note the range of the melting of the three mixtures and note the temperature of the complete liquefaction. Determine the mixed melting point of the organic compound and repeat

the determination and if the two as three determination do not check within

1 degree C do a third one.

Report:

Determination of melting point of pure

S.No	Compounds	Starting point (°C)	Finishing point (°C)

Exp. No: 4 Determination of boiling point of the unknown organic compound

Aim:

To determine the boiling point of the given unknown organic compound

Introduction:

Boiling point of a pure liquid is defined as the temperature at which the pressure on the liquid equal to the vapour pressure. The observed boiling point is directly proportional to the pressure. As the temperature raises the vapour pressure rises. It is necessary to state the pressure also while the boiling point for instant "boiling point 165°C |700mm". The standard boiling point is typically measured at atmospheric pressure, i.e 760mm of Hg presence of impurities influence the boiling point of the liquid and it depends on the nature of the impurity.

A non- volatile impurity may generally led to a sharp boiling point where as volatile impurities rises. The boiling point is determination of boiling point is useful for the identification of the pure liquid. A liquid as a rule will boil at a constant temperature are provided the pressure remaining constant. However, most mixture of liquid, boil over a fairly wide temperature even at a constant temperature.

A mixture of two different substance of the same melting point will show a melting point below that of each pure substance of the mixture. In contrast a mixture of two liquid of the mixture of the same boiling point as each individual one. Thus the boiling point is less useful for identification than the melting point.

Procedure:

Place 3-4 drops of the liquid whose boiling point is to be determined in an ignition tube immerse a capillary tube sealed at the other end in the liquid rises in the tube it means that it is not properly sealed. Attached the ignition tube to a thermometer by the means of the rubber band. Suspend the thermometer in the long neck flask containing paraffic oil in a tile tube. Heated the flask uniformly with a burner, until a rapid stream of bubble status coming out of the capillary tube

(because the air inside the tube warms and expands). At this point remove the burner and permits the flask to cold. The stream of bubbles become slower is searched when the babblings ceases and the liquid commences to rise in the capillary tube. This is the boiling point of the liquid.

Report:

The boiling point of the given organic compound is _____

Aim:

To recrystallize the organic and calculate the percentage of recovered benzoic acid.

Introduction:

The products of the chemical reaction can be impure. Purification of the products must be performed to remove by products and impurities liquids are customarily purified by distillation, while solids are purified by recrystallization. It is used to purify a solid. The process required a suitable solvent. A solvent is one which readily dissolves the solid (solute) when the solvent is hot but not when it is cold. The best solvent exhibits a large difference in solubility over a reasonable range of temperature. Example: water can be recrystallized solvent between 0-100°C hydrocarbon solvent such as hexanes or petroleum, ether have different temperature range since they can be cooled below zero degree but boil below 100 degree

Experimental procedure:

Using a weight paper, weight out about 1gm of impure benzoic acid for recrystallization and transfer it to a 125 ml conical flask. Add about 200 ml of distilled water, using on the hot plate, while stirring the mixture and boil gently to dissolve benzoic acid completely. Remove the flask from the hot plate and examine the solution. If these particles of the benzoic acid are still undisclosed, then add an additional amount of hot or cold water in a small increments and resume heating the solution. The objects is to dissolve the entire solid is only as much as hot or near boiling the yield of purified benzoic acid was be reduced. Keep adding water in small amounts until all of the benzoic acid is dissolved and the solution is boiling. If the solution is completely clear (through not necessarily colorless) and no solid benzoic acid is visible. Then add additional 10 to 15ml of water to the mixture and place the conical flask on a counter top where it will not be disturbed and cover with an upside small beaker. (To prevent dust contamination). Allow the flask to cool slowly will give the best shaped crystals after about 5 to 10 minutes. If the crystallization does not occur after 10 minutes, scrape the side of the flask about the level of the solution with the sharp end of a glass rod enough to audibly scratch the interior surface of the flask. This may dislodge some

undetectable, small crystals that are dropped into the solution and 'seed' the solution, helping through induce crystallization. A seed crystal can serve as a nucleation point for the crystallization process cooling the solution in the ice bath may also help at this point.

When the crystals have formed completely (may required in ice bath) collect the solid chemical by setting up a vacuum (suction) filtration on a properly fitted filter paper in the clean Buckner funnel apparatus. Pour the chilled mixture into a Buckner funnel, the water should filter quickly if not, check for vacuum leakage. Get all the crystals out of the flask using the spatula or stirring rod. Rinsing with one or two ml of cold water helps to get the crystals out of the flask and rinsing helps to remove impurities. Let the aspirator run for a few minutes to start the Buckner funnel, then press them as dry as possible on a large clean paper towel (hand dry), allow them to dry completely and transfer the dry sample to a pre-weighed weighing paper. Determine the weight dry crystals of required benzoic acid.

Recovery:

$$\text{Percentage of recovered sample} = \frac{\text{Weight of benzoic acid obtained after recrystallization}}{\text{Weight of benzoic acid recrystallization.}} \times 100$$

Result:

1. Sample name: Benzoic acid

2. Data of the impure Benzoic Acid:

a. Mass of the impure benzoic acid + weighing paper =

b. Mass of the weighing paper =

c. Mass of the impure benzoic acid =

3. Data of the recrystallized benzoic acid:

a. Mass of the recrystallized benzoic acid + weighing paper =

b. Mass of weighing paper =

c. Mass of recrystallized benzoic acid =

d. Calculate the percentage recovery =

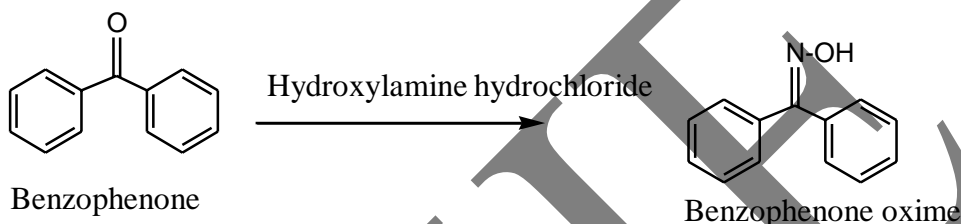
e. Melting point of the benzoic acid =

f. Structural formula for benzoic acid =

Exp. No: 6 Preparation of Benzophenone oxime from Benzophenone

Principle

When benzaldehyde is condensed with hydroxyl amine, one water molecule is removed and gives benzophenone oxime or benzophenoxime. This undergoes Beckmann rearrangement followed by hydrolysis gives benzanilide.



Requirements:

Chemicals	
Chemical Name	Quantity
Benzophenone	5 g
Hydroxylamine Hydrochloride	3 g
Rectified spirit	50 ml
Sodium hydroxide	8 g
Concentrated Hydrochloric acid	15 ml
Apparatus	
Apparatus Name	Nos
Round bottom flask (250 ml)	1
Reflux condenser	1
Wire-gauze	1
Glass beaker (250 ml)	1
Dropper	1
Glass rod	1
Water bath	1

Funnel	1
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Procedure:

A mixture of benzophenone, hydroxylamine hydrochloride, about 10 ml of water and rectified spirit is placed in a round-bottom flask. Sodium hydroxide pellets are then added in portions with constant shaking to the mixture in the flask. When the reaction becomes too vigorous the flask is cooled with tap water. When all the sodium hydroxide has been added, the flask is fitted with a reflux condenser and heated on a water bath for 15 minutes. The flask is then cooled, the contents poured into 100 ml of water containing conc.HCl. The precipitate is filtered at the pump, washed with water and dried in vacuum desiccators over sulphuric acid. (Expected yield: 5 g; Melting Point: 142°C)

Result:

1. Yield of the Product-
2. Melting Point of the Product-

Exp. No: 7 Preparation of Phenylbenzoate

Principle:

Phenol is treated with benzoyl chloride in presence of sodium hydroxide to get phenyl benzoate.

Requirements:

1. Phenol----- 1g
2. Sodium hydroxide -----1.5 g
3. Benzoyl chloride----- 2 ml

Procedure:

About 1 g of phenol is dissolved in 15 ml of sodium hydroxide solution in a conical flask. 2 ml of benzoyl chloride is added. Then the mouth of the flask is closed by a cork and shaken vigorously for 30 minutes. When the reaction is complete, the smell of benzoyl chloride will disappear. The precipitated phenyl benzoate is separated by filtration, washed with water, dried and the yield is noted. A little of the substance is recrystallized from alcohol, a sufficient excess of the solvent being used to prevent the separation of the material as liquid. Melting Point- 162°C

Result:

1. Yield of the Product-
2. Melting Point of the Product-

Aim:

To detect the extra elements present in the organic compound.

Procedure:

Sodium fusion extracts preparation

Melt a small piece of dry sodium in the small fusion tube. Add 0.1gm solid (or 3 drops liquid) to fusion tube. Heat gently at first then to red hottest. Quickly and carefully plug red heat end of the tube into 10 ml of distilled water in a china dish solution well a broken end of tube, boil and filter.

a. Test for nitrogen

1ml fusions extract and add few crystals of ferrous sulphate. Boil and cool add 2ml of dilute sulphuric acid solution blue or green solute or precipitate indicates the presence of nitrogen.

b. sodium nitroprusside test:

To 1ml fusion extract add 1ml nitroprusside solution violet color show the presence of sulphur.

Result:

1. _____ color solution is present.
2. _____ is present in the given organic compound.

Exp. No: 9 Separation of a mixture of two amino acid by ascending paper chromatography

Paper chromatography:

Paper chromatography is similar to the thin layer chromatography. In this technique a small spot of the sample is placed near one end of the strip of filter paper. The paper strip is suspended in a jar in such a way that the end of the filter paper is immersed in the developing solvent. The sample is separated into individual spots as the solvent ascends the paper in this case a distribution takes between water (absorbed by the filter paper an extent of 20%) the mobile solvent. If for this reason it is refer to as liquid-liquid partition chromatography. The different compounds may be identified to calculating the R_f values. Paper chromatography is useful for polar molecules like amino acids. The individual spots of amino acids are visualized by spraying cold ninhydrin solution and blue-violet color produced.

Most of the amino acid gives blue color (except protein which gives yellow color it indicates that the coloured product from is the same in the reaction) Paper chromatography is a valuable tool for the separation of 2 amino acid as they migrate at different rates.

Procedure:

From whattmann no:1 filter paper cut a strip measuring 30x10cm. Draw a line with a pencil about 3cm from one edge. Prepare amino acid solution by dissolving 120 mg each of glycine, protein, phenyl alanine, leucine and aspartic acid in the 20 ml of water. Spot the paper with the mixture of the amino acid and also spot each of the above amino acids about 1.5 apart the spot should be 2 to 3 mm. If it is too large it may led to poor resolution dewing development. Allow and insert it into a glass cylinder containing the developing solvent (n-butanol, glacial acetic acid, water respectively in the rate 4:1:5) make sure the paper dose not touch the sides of the cylinder and the sample spots are above the level of the solvent. The solvent will rise by capillary action when the solvent has migrated to third the length of the paper remove the paper and mark the position of the solvent. Dry the chromatography now sprays it lightly but evenly with ninhydrin solution and dries it again. Heat the chromatography in an oven at 105 degree C for 15 minutes,

where by the colors are visible mark the positions of the spots with a pencil and estimate the R_f value.

Result:

The R_f values of the given amino acid mixture are = _____

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Exp. No: 10 Separation of a mixture of two sugars by ascending paper chromatography

Paper chromatography:

Paper chromatography is similar to the thin layer chromatography. In this technique a small spot of the sample is placed on one of the strips of the filter paper. The paper strip is suspended in a jar in such a way that the end of the filter paper is immersed in the developing solvent. The sample is separated into individual spots as the solvent ascends the paper. In this case a distribution takes place between water (absorbed by the filter paper to be extends of 20%) and the molecule solvent for this reason. It is also called as liquid-liquid partition chromatography. The different compounds may be identified by calculating R_f values. Paper chromatography is useful for polar molecules like sugar. The individual spots of sugar are visible by spraying with mixture of KMnO_4 and Na_2CO_3 solutions.

Procedure:

From whatmann paper no:1 filter paper cut a strip measuring 30x10 cm. Draw a line with pencil about 3cm from one edge. Prepare the sugar solution by dissolving 120gm of each of fructose and xylose in 20ml of water. Spot the paper with the mixture of the sugar and allow put a spot and each of the individual about 1.5cm apart. The spot should be 2-3 mm in a diameter if it is too large it may lead to a poor resolution during development. Allow the spot to dry. Fasten the paper with dry and insert it into the glass cylinder containing the developing solvent (n-butanol, glacial acetic acid) and water respectively in the ratio 4:1 make sure paper does not touch the side of the cylinder and the sample spots are above the level of the solvent that has migrated two-third the length of the paper and mark the position of the solvent. Dry the chromatography now spray it lightly but evenly with KMnO_4 and Na_2CO_3 solution and dry it again that the chromatography in an oven at 105°C for 15 minutes. When dry the colors are visible mark the position of the spot with a pencil and estimate the R_f values.

Result:

The R_f values of the given sugar mixture = _____