Biology Chromatography Investigation Results

Table of results to show RF values of amino acids found in egg white following dissection by trypsin.

RF value = Distance moved by spot
Distance moved by the solvent

Sample	Cm Distance moved	Cm moved by solvent	RF Value	Identity of amino
Alanine	4.9	16.3	0.30	
Leucine	11	16.3	0.67	
Egg white A)	5.2	16.3	0.31	
B)	11.2	16.3	0.68	

RF table

Amino acid	RF Value
Lysine	0.14
Arginine	0.20
Aspartic Acid	0.24
Glycine	0.26
Serine	0.27
Glutamic Acid	0.30
Threonine	0.35
Alanine	0.38
Proline	0.43
Tyrosine	0.45
Methionine	0.55
Valine	0.60
Phenylalanine	0.68
Isoleucine	0.72
Leucine	0.73

Class Data

Sample	1	2	3	4	5	6
Alanine	0.3	0.3	0.3	0.3	0.3	0.3
Leucine	0.7	0.7	0.7	0.7	0.7	0.7
Egg white A	0.3	0.3	0.3	0.3	0.3	0.3
B	0.7	0.7	0.7	0.6	0.6	0.7

Get a piece of chromatography paper and draw a pencil line 4cm from the bottom. Dispense the egg white, alanine and leucine equally spaced along the line with a micropipette. Pour solvent in bottom of jar up to 1cm place paper in jar ensuring the solvent doesn't cover the pencil line (line of origin). Place lid on and leave in fume cupboard for 8-16 hours. Draw a line were solvent reached. The amino acids should show as purple spots. To calculate the RF values divide the distance traveled by amino acids by the solvent front.

On the chromatography paper there were a few faint spots that could be other amino acids present or chemicals from the sweat off our fingerprints.
The RF values of the amino acids in the egg white are A) 0.31,B) 0.68 this shows according to the RF table on page 1 A = Glutmatic acid and B = Phenyalanine.
The class data was very consistent in that every body got the same RF values. In the class the distance moved by the solvent (1% Ninhydrin in butan-1 OL) carried between 14cm and 16.3cm but no matter how far the distance the solvent moved the amino acids would move correspondingly and will not change the RF values.

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There are factors that can affect the RF value. Proteins in the albumen solution not being fully broken down. There still may be some peptide bonds holding the amino acids

together which would mean that once put in the solvent the proteins would extract water from the solvent which would cause other small spots on the paper. Leaving the trypsin enzyme for longer in the albumen to complete the hydrolysis could make it more accurate. The fibres in the paper differ in size and shape, which naturally limits the precision and means that the spots are not as clear.

You could leave it as long as you liked as long as you left enough time for the proteins to be broken down it is irrespective how far the distance the solvent moves the amino acids would move correspondingly and will not change the RF values.

There are many errors that could occur during this experiment, which could affect the results. The amount of the solutions dispensed on the line of origin should be measured more accurately because the micropipettes are difficult to handle. This ensures enough and equal concentration of amino acids thus resulting in more accurate readings and enhanced RF values.