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FINAL YEAR PROJECT 'PAPERFUGE' (HAND-DRIVEN CENTRIFUGE)

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DECLARATION

We hereby certify that this submission is our own work towards the award of a Bsc degree, and that, to the best of our knowledge it contains no material previously published by another person nor material which has been accepted for the award of any other degree of the university, except where due acknowledgement has been made in the text.

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ABSTRACT

Malaria is a common disease in Ghana affecting 3.5 million people every year. [1] To diagnose malaria in patients, blood samples are needed. In order to prepare these samples and isolate the malaria parasites, a centrifuge is needed.

But commercial centrifuges are expensive with an approximate price of 1200 to 2000 Ghana cedis. Not to mention they are electrically powered.

Thus, a solution was engineered inspired from an ancient toy dating back to 3300 B.C. called a whirligig toy[2] to make an low-cost, lightweight and human-powered paper centrifuge which they called a 'paperfuge'.[3]. Experiments were conducted to determine factors that affect the spinning speed of a hand powered centrifuge, to know which factors best increase the speed. And to isolate malaria parasites for diagnoses in less than 15 minutes.

Our designs model of the paperfuge achieved highest speeds of 58,000rpm in 1 minute. We used this speed to separate pure plasma from whole human blood. With over 78 experiments conducted with whole blood, the average speed was 36,875rpm under 0.7secs. These experiments were used to detect malaria parasite in blood samples.

DEDICATION

We dedicate this work to our family and loved ones.

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ACKNOWLEDGEMENT

No matter what man does, he cannot count the favors of the Almighty God. It is always through His Grace, Mercy and blessings that we were able to undertake this project and complete this piece of work.

We are indebted to our lecturer and supervisor, Mr. Prince Odame, for his support and guidance throughout our time in KNUST and this project work.

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CHAPTER 1

INTRODUCTION

1.1 Background of Study

Centrifuges are vital in health care settings; they are used for blood tests and analysis to diagnose many diseases. A lot can be deciphered from a person's blood sample. Malaria is a common disease in Ghana. Diagnosing malaria requires blood samples to be taken from a patient for lab testing. In order to prepare these blood samples, a centrifuge is needed. The centrifuge separates plasma from whole blood or isolates the malaria parasites. Sample preparation which includes separating plasma from whole blood or isolating parasites is quite a challenge since it is the first step in diagnostics. [2]

A centrifuge is simply a machine that spins around to make a large and useful force. Small centrifuges in scientific laboratories are used to for example, separate blood products.[4]

Unfortunately, not every area in Ghana can afford centrifuges and neither do they have electricity to power these centrifuges. Thus, a solution was engineered by Stanford inspired from an ancient toy dating back to 3300 B.C.E called a whirligig toy[2] to make an low-cost, lightweight and human-powered paper centrifuge which they called a 'paperfuge'.[3] The paperfuge reaches the speed of 125,000rpm and an equivalent centrifugal force of 30,000g. It can separate pure plasma from whole blood in less than 1.5minutes and it can isolate the malaria parasites in 15minutes.[5]

1.2 Problem Statement

Commercial centrifuges are expensive with an approximate price of 1200 to 2000 Ghana cedis. They are electrically powered. Not all areas of Ghana can afford centrifuges or even have electricity to power these machines. According to an analysis, 82.5 percent of the population in Ghana had electricity by 2016[6]

1.3 General Objective

This project focuses on implementing the paperfuge in Ghana for areas with no access to electricity to power centrifuge machines.

1.4 Specific Objectives

- 1. The aim is to reduce diagnostic time by increasing the spinning speed of the paperfuge.
- 2. It is also to determine the factors that affect the spinning speed of the paper centrifuge.
- Another aim is to determine which factor(s) best increase the spinning speed of the paper centrifuge.
- 4. In the end, to isolate the malaria parasite from the blood plasma in less than 15minutes for diagnoses.

1.5 Significance of the Work

- 1. This work is useful in health care facilities for diagnostic purposes.
- 2. This work is useful for areas with no electricity.
- 3. This work is useful for health facilities that cannot afford commercial centrifuges.

CHAPTER 2

LITRATURE REVIEW

2.1 History of Centrifugation

Centrifugation is traced back to the 15th century when they were hand-driven. These early hand-driven centrifuges could reach speed of up to 3000rpm. In those early years, centrifugation was used for non-biological purposes like separating milk and collecting precipitates. Back in 1864, a system of milk separation was introduced by Antonin Prandtl which separated cream from milk.[7]

The prospect of centrifugation in the laboratory was introduced by Friedrich Miescher. In 1869, Miescher used a crude centrifuge to separate a cell organelle. This later led to the discovery of biological constituents, later to be known as nucleic acids.[7]

By 1950, centrifugation of tissue fractionation, a process developed by a plant virologist, Myron K. Brakke [7].

In 1954, Beckman Coulter purchased spinco, forming the basis of spinco centrifuge division. He quickly improved he design of the centrifuge most of which are still used today. For example, he upgraded it by substituting high speed motors and also upgraded the material for the rotor blade. The original rotors such as those built by Svedberg were made of tensile steel. Today, materials such as titanium and aluminum alloys are used to withstand high centrifugal forces. Vacuum systems have been added to reduce friction and control temperature too. [8]

Beckman went on to make ultracentrifuges. This company is known as Beckman Coulter and is still a major manufacturer of centrifuges used today. [7]

2.2 Types of Centrifuges

A few kinds of centrifuges are mentioned:

- Small bench centrifuges
- Large capacity refrigerated centrifuges
- Ultra centrifuges
- Fixed angle rotors

2.3 Principle of Centrifuge

Basically, a centrifuge spins liquid at high speeds which creates a strong centripetal force causing denser materials to settles towards the bottom of the centrifuge tube more rapidly than they would under the normal force of gravity. [4]

A liquid which contains suspending particles of homogeneous density streams through a conical tube, the velocity is always at it greatest at the apex of the cone and decreases at the base. When the tube containing the homogeneous liquid is placed in the centrifuge with the apex pointed away from the centre of rotation, the particles acquire, in addition to the velocity owing to the liquid stream, a velocity which goes in the opposite direction owing to the centrifugal effect.[9]

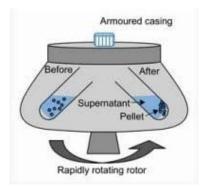


Figure 2.1: Centrifugation Theory [10]

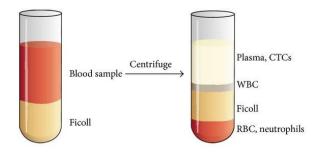


Figure 2.2: Principle of Density Centrifugation[11]

2.4 The Modern Centrifuge

A typical modern centrifuge can speed up to 70,000 rpm. The size of the tubes and the speed vary from rotor to rotor. Generally, the centrifuge spins at 3500 rpm for 10 to 15 minutes. It has a compartment than spins on its axis. A centrifuge rotor is the rotating unit of the centrifuge which has fixed holes at an angle where the tubes are placed. [12]



Figure 2.3: A Modern Centrifuge [13]

2.5 What Is The Human-Powered Centrifuge?

Dr. Prakash from Stanford University was inspired by the whirliging toy to develop a humanpowered centrifuge with low cost. He visited a rural area in Uganda and saw they had used a centrifuge as a doorstop in their local clinic due to the fact that there was no electricity to power the centrifuge. Dr. Prakash then built a human-powered centrifuge by attaching capillary tubes to a plastic-coated paper disk and used strings attached to the disk to create an oscillatory force [14]

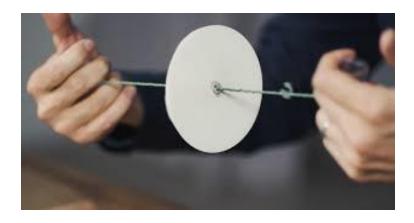


Figure 2.4: Paperfuge[15]

He described the paperfuge as a non-linear, non-conservative oscillator system. Presently it goes at a speed of 125,000rpm with an equivalent centrifugal force of 30,000g. This speed was used to separate pure plasma in less than 15minutes to isolate malaria parasites in 15 minutes from the human blood. [16]

Basically, the paperfuge consists of successive 'unwinding' and 'winding' phases. During the unwinding, the outward input force which is applied by human hands on the handles accelerates the disc to maximum speed. Through the winding phase, the input force falls to zero, allowing the disc's inertia to rewind the strings and draws the hands back inwards. Due to the fact that the strings are flexible, they wind outside the geometric zero-twist point. Then, after arriving at a tightly paced super coiled state, the motion of the disc comes to a momentary halt. At this position, an outward force is re-applied, which unwinds and winds the strings. This cycle repeats itself as shown in figure 2.5. [16]

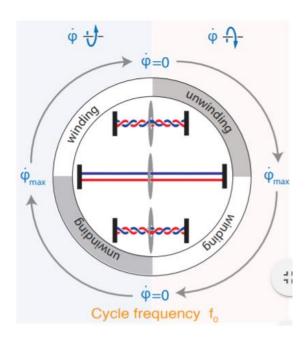


Figure 2.5: Mechanism of Paperfuge[2]

The figure below shows high speed images of the rotation of the hand powered centrifuge. (i) Shows the wound state, (ii) shows the unwound state and (iii) shows the final wound state for half a cycle. It shows the super coiling of the string during the wound states.[2]

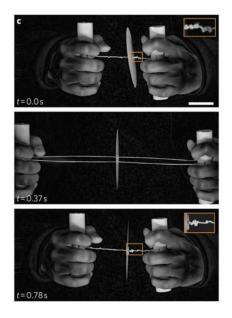


Figure 2.6: Rotation of Paperfuge[2]

The figure below shows a graphical representation of the radius of the disc and it corresponding revolution per minute. It shows a plot of φ_{max} and f_o for different discs (radius = [5, 12, 25, 50, 85] mm) of the hand powered centrifuge. From this graph it can be seen that the radius of the disc influences the speed achieved by the disc. The smallest disc of radius 5mm reached a speed of 125,000rpm while the disc of radius 50mm reached below 50,000rpm. [2]

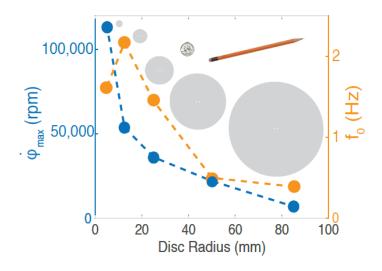


Figure 2.7:Graph of Disc Radius against RPM[2]

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Egg beaters and salad spinners were also tried but it was not successful due to low spin-rates. But the paperfuge has been a success. The smallest disc, of radius 5mm reached peak angular speeds of 125,000 rpm using only human power. The disc with a 50mm radius reached 20,000 rpm. [17] The toy is described at a non-linear, non-conservative oscillator: in every rotation cycle, the input energy is introduced by human hands which are the applied force and it is dissipated by the system through air drag and in the strings. [16]

Mathematically, Newton's law states that the rate of change of angular momentum is equal to the total torque or turning effect. Where I is the moment of inertia, ϕ is the angle of the disc and the τ -term denotes components of torque.[17]

$$I\phi = \tau_{input}(\phi) + \tau_{drag}(\phi) + \tau_{twist}(\phi)$$

Three turning effects, one due to the manual input force applied by the operator, second is the twisted state of the string and then third is due to the air drag on the disc all affect the spinning.

[17]

2.5.1 Material and Method Used

The paperfuge is composed of a two card-stock paper discs. A fishing line is used for the strings to provide high tensile strength. For the handles common wood or PVC pipe is used. Drinking straws were used to create safe and easy mounting of the capillaries. The straws were sealed to serve as a safety measure incase the capillary leaks. Two holes were poked into the center of two equally cut circular shaped discs. During the spinning, friction occurs between the paper and the string and to avoid this, acrylic ovals were used so that the holes on the acrylic align with the holes on the disc. The pipe is wedged between the two disks mounted radially, and the device is spun for a short time. And the disks are attached using Velcro. [16]

The image below shows the materials used.

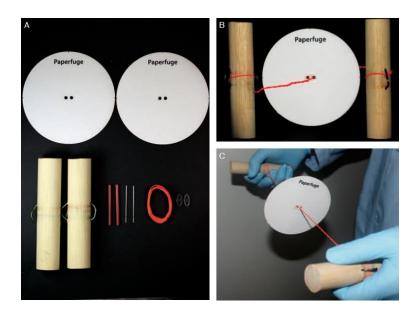


Figure 2.8: Materials of Paperfuge[18]

As a safety measure, the capillary tubes are made of plastic to avoid it from shattering. Then the capillary tubes are put into pipes to avoid accidental leaks. Also, the two disks covering the pipe are to avoid the exposure of the blood to the user. [16]

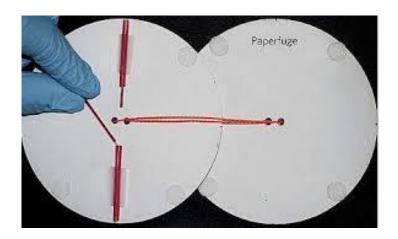


Figure 2.9: Capillary Tubes Mounted on Paperfuge[19]

CHAPTER 3

METHODOLOGY

This chapter discusses the process and procedure that was used to design the hand powered centrifuge and increase the speed of the hand powered centrifuge. The experimental design for this research is acquired from these experiments.

3.1 Materials

Fishing net and twine

Button

Card board

Capillary tubes

Holders

Tachometer

3.2 Experimental Design

This subheading includes the preliminary and actual experiment

3.2.1 Preliminary Experiment

In the first experiment (figure 3.10), a shoe maker thread was used and hard cover of an exercise book was cut into disc with a radius of 2cm. This was spinning at a speed of 35,000rpm.

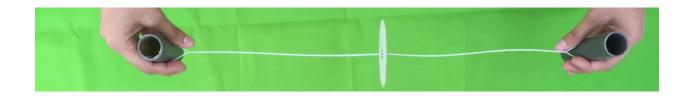


Figure 3.10: Experiment 1

In the second experiment (figure 3.11), just a button was spun but with wool thread. The thread tore apart in less than 5 minutes, unable to withstand the force.



Figure 3.11: Experiment 2

In the third experiment (figure 3.12), a disc of 5cm radius with holes 3cm apart was used this turned out to be slow in the spinning process. From this, it was concluded that for the disc to spin, the center of mass should be close to the center. The further away the holes from the center, the distance from the center of mass of the disc and hole increases. Hence, the effort will not act through the center of mass of the disc and the word done will also include distance between holes and center of mass of disc. This will reduce the velocity unlike when the holes are at the center of mass of the disc.

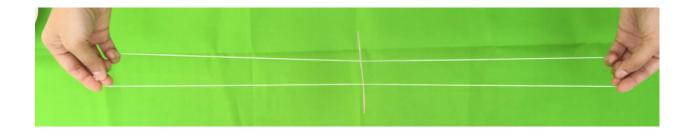


Figure 3.12: Experiment 3

In the last, fourth experiment (figure 3.13); two threads were looped into 4 holes instead of one thread looped through 2 holes to try increasing the speed. There was an obvious increase in the speed. Torque is proportional to acceleration, so with twice the number of holes there will be twice the input and twist torques. This will result in twice the acceleration and so twice the speed.

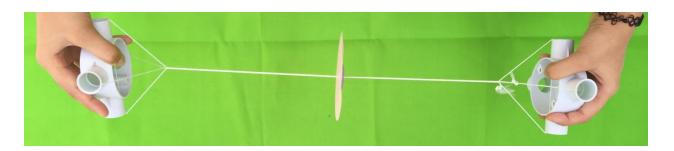


Figure 3.13: Experiment 4

An elastic string was also used on a button but it did not spin due to the elasticity of the string which led to the belief that that the string must be taut for the disc to spin (figure 3.13).



Figure 3.14

A table of the preliminary experiments is shown below.

Table 1: Table of Experiments

	Thread (type)	Disc (radius	Number of	Speed (rpm)
		in cm)	holes in disc	
Experiment 1	Shoe maker	2.5	2	35,000rpm
	thread			
Experiment 2	Woolen thread	1.0 (button)	2	Thread tore
Experiment 3	Fishing net and	5.0	2 (3cm apart)	Did not spin
	twine			
Experiment 4	2 sets of fishing	5.0	4	40,000rpm
	net and twine			

(Source: Preliminary Experiment Analysis, 2018)

The number of holes and threads could be increased to for example 6 or 8, but, that would make the design complicated and difficult to hold as well.

3.2.2 User Safety Measures

To prevent exposure to blood gloves were worn before beginning the experiment. Secondly, the capillaries were mounted onto sealed straw holders that contain any accidental leaks. Also, the paperfuge had two discs, one with the straw capillary holders and one disc acting as a cover held together using the Velcro strips. By covering the disc, exposure to blood is minimized.

3.2.3 Experiment on Water and Chalk

For the actual experiment, the design from experiment 4 was used (table 1). Two straws of 4.5cm were cut and one end of them was sealed with epoxy and left to dry for about 10 minutes. The straws act as holders for the capillary tubes. The open end of the straw was placed toward the center and the closed end toward the outer part of the disc. The straws were placed opposite each other to enable the paperfuge to be balanced due to the fact that it helps increase its spinning speed. The holders also act as a safety measure to prevent blood from spilling incase of leakage.

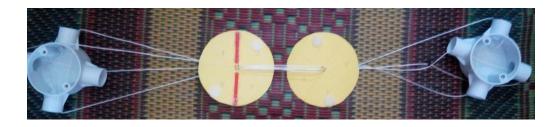


Figure 3.15: Paperfuge with handles at both ends

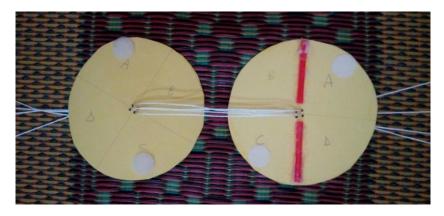


Figure 3.16: A red straw as holders and left disc covers the right disc

A solution of powdered chalk and water was mixed and filled into the capillary tubes. Both ends of the capillary tubes were sealed with a drop of epoxy. Then the tubes were left to dry the epoxy for about 10 minutes before placed into the holders. The mixture was spun for 2minutes and it was seen that the chalk particles had settled at the bottom of the mixture.



Figure 3.17: Chalk particles suspended in water



Figure 3.18: Chalk particles settled to the far right

This phenomenon is based on the principle of centrifugal and centripetal force. The centrifugal force keeps the disc in circular motion whiles the centripetal force is directed toward the center of the disc. The centrifugal force acts on the fine solid particles in suspension in the liquid. These chalk particles are pushed to the bottom of the capillary tubes. The magnitude of the centrifugal force 'F' depends on the speed 'v' of rotation, the mass 'm' of the fine particles and the radius 'r' of the disc. This force is mathematically described as

$$F = \frac{mv^2}{r}$$

3.2.4 Experiment on Whole Blood

To test the paperfuge with whole blood, blood samples were acquired from Ahmadiyya Muslim Mission Hospital, Asokore-Ashanti. 78 blood samples with positive malaria and negative were

used. All precautionary measures were undertaken in handling blood samples. A total of 78 experiments were done on said blood samples by placing two samples at a time on the paperfuge.

First of all blood samples were infused into hematocrit capillary tubes (40mm long) through the action of capillarity. Both ends of capillary tubes were sealed with epoxy and left to dry for 20 minutes. After making sure capillary tubes were sealed, two tubes were mounted onto the paperfuge opposite each other. The paperfuge was then spun and separation of blood particles was seen.



Figure 3.19: blood samples in capillary tubes sealed at both ends with epoxy 3.2.5 Malaria Testing

Due to the unavailability of Quantitave Buffy Coat (QBC) capillary tubes and a florescent microscope, another method to detect malaria parasite in centrifuged blood was adopted. The buffy coat achieved from centrifugation earlier on was put on a glass slide and smeared as the same method for thick blood film with geimsa stain. The plasma was left in the capillary tube. The slide was then placed under electronic microscope for viewing. With the help of lab technician present, malaria parasite was seen through the microscope.

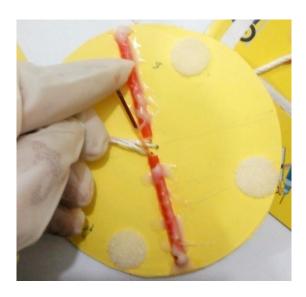


Figure 3.20: mounting capillary tubes on paperfuge



Figure 3.21: measuring speed of paperfuge while spinning sample

3.2.5.1 Geimsa Stain Method

In the geimsa staining method, the buffy coat extracted from the centrifuged blood was spread on a clean glass slide and left to dry. After it was dried up, geimsa stain was added on to the sample and left to 10 minutes. Afterwards, the geimsa stain is washed with a buffer solution to remove the geimsa stain from the slide. It was dried again. Lastly, after it has dried, oil immersion was spread on the sample and put under electronic microscope for viewing.

The features that distinguish the malaria parasite are that it has a blue colored cytoplasm and pink or purple or red colored chromatin dots as well as seen in fig 3.22 below.



Figure 3.22: glass slides prepared for viewing under microscope

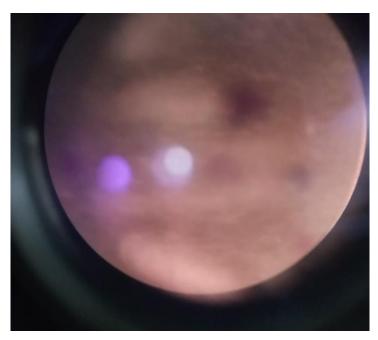


Figure 3.23: malaria parasite sighted under microscope

CHAPTER 4

RESULTS AND DISCUSSION

4.1 Chapter Summary

This chapter comprises the results of testing the paperfuge with 30 blood samples, their corresponding speeds and the time it took to separate plasma from whole blood.

4.2 Results of Testing The Paperfuge

The focus is to separate blood plasma from whole blood in less than 1.5 minutes and being able to also detect malaria parasite in the blood. Thereby proving the paperfuge can be used to replace a commercial centrifuge.

Average speed was 36,875rpm under 0.7 seconds (0.0116667 minutes). The table of results is attached in the appendix. Due to unavailability of QBC capillary tubes and florescent microscope, the serum from separated blood sample was used. Malaria was detected using the geimsa stain method from the serum.



Figure 4.24: slight separation achieved after 45secs

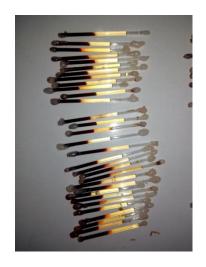


Figure 25: very clear separation after a minute of centrifugation

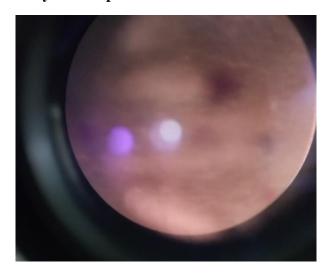


Figure 4.26: Purple dots representing malaria parasite from microscope lens
4.3 Comparison Between Commercial Centrifuge And Paperfuge

Table 2: comparison between commercial centrifuge and paperfuge

Centrifuge	Speed(rpm)	Time(min)	Paperfuge	Speed(rpm)	Time(min)
Malaria +ve	4,000	15 min	Malaria +ve	40,000	1.00 min
Malaria +ve	4,000	15 min	Malaria +ve	30,000	0.75 min
Malaria +ve	4,000	15 min	Malaria +ve	45,000	0.75 min
Malaria -ve	4,000	15 min	Malaria -ve	30,000	0.75 min

Malaria -ve	4,000	15 min	Malaria -ve	40,000	1.00 min
Malaria -ve	4,000	15 min	Malaria -ve	45,000	1.00 min
Malaria +ve	4,000	15 min	Malaria +ve	30,000	2.00 min

From the preliminary experiment, the design with four holes and two threads was used to try increasing the speed. There was an obvious increase in the speed. Torque is proportional to acceleration, so with twice the number of holes there will be twice the input and twist torques. This will result in twice the acceleration and so twice the speed.

From the table above, it was concluded that the paperfuge is a simpler design with high speed and just as efficient as the commercial centrifuges. But, in terms of speed, the paperfuge is many rpm's higher than the commercial centrifuge. The paperfuge takes considerably less time as the commercial centrifuge.

The materials used to design the paperfuge are cheap and readily available in the market.

In the case of power outages in hospitals, laboratory work can progress smoothly due to the fact that this design requires hand power. Also, areas with no access to power can easily use this paperfuge for centrifuging blood samples.

CHAPTER 5

CONCLUSION

5.1 Chapter Summary

This chapter comprises the results of testing the paperfuge with blood samples and detecting malaria parasite after centrifugation.

5.2 Conclusion

We were able to establish the factors that affect the spinning of the paperfuge. We were also able to determine how to increase the paperfuge speed. We were able to achieve separation of blood particles in 1 minute. And finally detect the malaria parasite in the Buffy coat achieved after centrifugation. Some studies suggest that diagnoses by direct centrifugation appear to be more sensitive as usual microscopy. [20]

5.3 Limitations

Due to the unavailability of the QBC capillary tubes and florescent microscope another approach was taken. Also, plastic capillary tubes were unavailable so glass capillary tubes were used. This resulted in spinning the paperfuge a few 1000 rmp's less to avoid the shattering of the glass tubes.

5.4 Recommendation

It is recommended to use a QBC capillary tube which eases the task of observing malaria parasite under the florescent microscope. It also essential to use plastic capillary tubes to avoid shattering of the glass tubes during high speed spinning of the paperfuge.

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APPENDICES

APPENDIX 1: TABLE OF RESULTS

 Table 3: Table of results for actual experiment

Experiment	Speed (rpm)	Time (mins)	Malaria (+ve/-ve)
1	40,000	1.00	+ve
2	30,000	0.75	+ve
3	45,000	0.67	+ve
4	30,000	0.75	-ve
5	40,000	1.00	-ve
6	45,000	1.00	-ve
7	30,000	2.00	-ve
8	40,000	0.83	+ve
9	45,000	0.75	+ve
10	46,000	0.50	+ve
11	45,000	0.67	-ve
12	47,000	0.67	-ve
13	48,000	1.00	+ve
14	40,000	1.00	+ve
15	30,000	0.50	-ve
16	35,000	0.50	+ve
17	41,0000	0.50	+ve
18	40,000	0.67	+ve
19	30,000	0.67	+ve

20	35,000	0.83	-ve
21	40,000	0.50	-ve
22	45,000	0.67	-ve
23	46,000	0.83	+ve
24	47,000	1.00	+ve
25	48,000	1.00	-ve
26	48,000	0.67	+ve
27	45,000	0.67	+ve
28	40,000	1.00	+ve
29	40,000	1.00	-ve
30	30,000	0.67	-ve
31	35,000	1.00	+ve
32	38,000	0.83	-ve
33	40,000	0.83	-ve
34	45,000	0.83	-ve
35	48,000	1.00	+ve
36	46,000	1.50	-ve
37	35,000	0.67	+ve
38	40,000	0.67	-ve
39	45,000	0.83	+ve
40	46,000	1.00	-ve
41	47,000	0.83	+ve
42	48,000	1.00	+ve

43	49,000	0.67	+ve
44	50,000	0.67	-ve
45	51,000	1.00	-ve
46	52,000	1.00	-ve
47	53,000	1.00	-ve
48	54,000	1.00	+ve
49	49,000	0.67	+ve
50	52,000	0.67	+ve
51	51,000	1.00	+ve
52	50,000	1.00	+ve
53	49,000	0.83	-ve
54	48,000	1.00	-ve
55	40,000	1.00	-ve
56	49,000	0.83	+ve
57	52,000	0.83	+ve
58	40,000	0.83	-ve
59	45,000	1.50	-ve
60	49,000	1.50	-ve
61	48,000	0.83	+ve
62	46,000	0.83	+ve
63	50,000	0.83	+ve
64	45,000	1.00	-ve
65	45,000	1.00	-ve

66	48,000	1.50	+ve
67	40,000	1.00	+ve
68	42,000	0.67	+ve
69	40,000	1.00	+ve
70	30,000	0.75	+ve
71	30,000	0.67	+ve
72	45,000	0.67	-ve
73	30,000	0.75	-ve
74	40,000	1.00	-ve
75	45,000	0.67	+ve
76	30,000	1.50	+ve
77	35,000	1.00	-ve
78	45,000	0.75	+ve
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