



RESEARCH ARTICLE

DETECTION AND COMPARISON OF NORMAL AND MENSTRUAL BLOOD SAMPLES FOUND AT CRIME SCENE

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ABSTRACT

Body fluids are the common and important type of forensic evidence. In particular the identification of Menstrual (liquid or stain) is often a key step during the investigation of rape case. This paper presents a method which is simple and easy for the differentiating menstrual blood from normal blood through its composition. In this microscopic analysis, hemoglobin content, RBC and WBC count was done as a preliminary study to distinguish the type of blood samples. The attempt has been made to determine the blood group from liquid menstrual blood by using antiserum ABH. Forensic analysis was done by performing confirmatory test of normal blood using Teichmann reagent which gives negative result with menstrual blood can be a good parameter for differentiating the menstrual blood from normal blood.

Key words: Menstrual blood, Hemoglobin, Stain, blood.

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INTRODUCTION

Menstrual blood is a complex biological fluid composed of blood, vaginal secretion and the endometrial cell of the uterine wall as they exist immediately prior to menstrual (Fraser *et al.*, 1985) Menstrual blood is not highly oxygenated, its darker than normal blood. It is basically a waste product contain dead and no longer functional tissue (Kao *et al.*, 2003; Strowitzki *et al.*, 2006; DeSouza *et al.*, 2005). The concentration of Iron, Hemoglobin and protein is less in menstrual blood than in normal blood. Sexual assault is unique among crimes as it leaves distinct physical evidence. In these types of cases it is necessary to distinguish that the stain was caused by a hymen rupture or menstrual fluid while it may be impossible to distinguish the source of the blood by visual inspection only. The Difference will be found in the ratio of certain blood cells and the presence of tissue parts that are not normally found in peripheral blood as well as other cellular differences. In past different type of techniques was used like latex agglutination tests for fibrin fibrinogen degradation production in the forensic identification of menstrual blood and identification of menstrual blood by simultaneous determination of FDP-D dimer and myoglobin content (Akutsu *et al.*, 2011) and Raman spectroscopy coupled with advanced statistics for differentiating menstrual and peripheral blood (Aliaksandra *et al.*, 2012) and other body fluids on various substrate using ATR FT-IR spectroscopy and reported the difference between

peripheral blood and menstrual blood by protein and enzyme named plasminogen (Alicia *et al.*, 2016). Now, in the present study we are using a simple technique to differentiate the menstrual blood from normal blood by detection the variance in the number of cells in both menstrual and normal blood of same individuals and Teichmann test is used for determination of blood and antiserum is used for the determination of blood group.

MATERIALS AND METHODS

Material: chemical reagents including teichmann reagent (Potassium chloride 0.1%, Potassium iodide 0.1%, Glacial acetic acid 10ml), Hayem's fluid, Turk's solution, Sahli's Haemoglobinometer and Hemocytometer grid, HCl, distilled water and antiserum ABH is used for the blood group detection

Sample collection: : 20 normal females subjects were being selected with age group between 18-25yrs. to give the menstrual and normal blood samples. The Liquid menstrual blood samples collected in the sample container and stained menstrual blood samples collected in the airtight or zip locked plastic envelopes. In case of normal blood sample approximately 2ml blood was collected from same individuals.

Method: Menstrual and normal blood are taken from same individual and preliminary study was done for distinguish the type of blood samples. haemoglobin level was determined by using sahli's haemoglobinometer in this anticoagulated blood is added to the 0.1 N HCL and kept for 5-7 minute to form acid haematin. The colour of this acid haematin was matched with

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the solution present in the calibration tube. Distilled water is added to the acid haematin until the colour matches and then notices the readings of both samples (International Council for Standardization in Haematology, 1996). Then RBC and WBC cell are count with the help of Neubauer's Haemocytometer slide in both samples of same individual and the attempt has been made to determine the blood group from menstrual blood samples by using simple antisera ABH. We can detect the blood group from menstrual blood by using antiserum ABH (Ghai, 2008)

Microscopic analysis: Microscopic analysis has been done by making thin smear of blood and used for differentiate the menstrual blood from normal blood samples and microscopy is very good parameter for differentiating the menstrual blood from normal blood. The PH of the menstrual blood samples, with the help of pH paper, pH of blood samples was observed and observation was taken at different interval of time.

Forensic analysis: Forensic analysis was done by performing confirmatory test with menstrual blood, Teichmann test was performed on the menstrual blood in this teichmann solution is add on the blood sample and cover with the cover slip and then warm the slide on 65°C for 10 -20 seconds and then allow it to cool and observe under microscope.

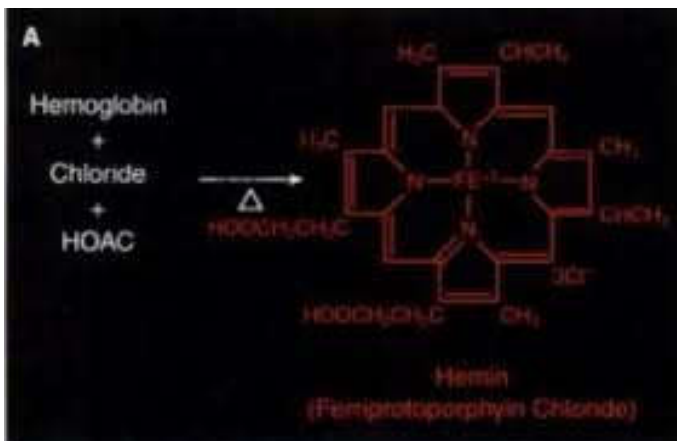


Figure 1. The hemoglobin reaction with Teichmann solution.

RESULTS AND DISCUSSION

The preliminary study that is to distinguish the blood samples like Haemoglobin, RBC count and WBC count show the great variation. In this we observed that level of hemoglobin, RBC and WBC is very less in menstrual blood as compare to the normal blood samples. Normally the value of hemoglobin in normal blood samples consider is 10-16gm% and in menstrual blood is from 2-5gm%.

Table 1. Show the different parameter have different values present in the normal and abnormal blood samples of the same individuals

Sr.No.	Parameter	Normal blood samples	Abnormal blood samples
1.	Hb	10-13gm/dl	2-5gm/dl
2.	RBC	4-5million/cm	2-3million/cm
3.	WBC	5-8cu/mm	4-6cu/mm

In this study find that hemoglobin level in menstrual blood is very low as compare to the normal blood samples (shown in Table 1). According to results it gives a 90% positive results in

20 blood samples of female individuals. Similar results for haemoglobin concentration in normal blood to determine the presence of insufficient red blood cell mass to adequately deliver oxygen to peripheral tissue (Andrew *et al.*, 2003). And the count of RBC in normal blood samples are between 4-5million/cm but in menstrual blood samples the range of RBC count approximately between 2-3million/cm (shown in Table: 1). According to our observation the value of RBC cells in menstrual blood is very low. Number of white blood cells that we observed in our results are between 5-8cu/mm in case of normal blood samples but in menstrual blood samples the number of WBC cell is very low that is observed between 4-6cu/mm (as shown in Table 1). The author has studied a method for rapid and simultaneous analysis of nucleated red blood samples. Main method and device for the simultaneous and quantitative, flow cytometric analysis of nucleated red blood cells and white blood cells from whole blood cells.

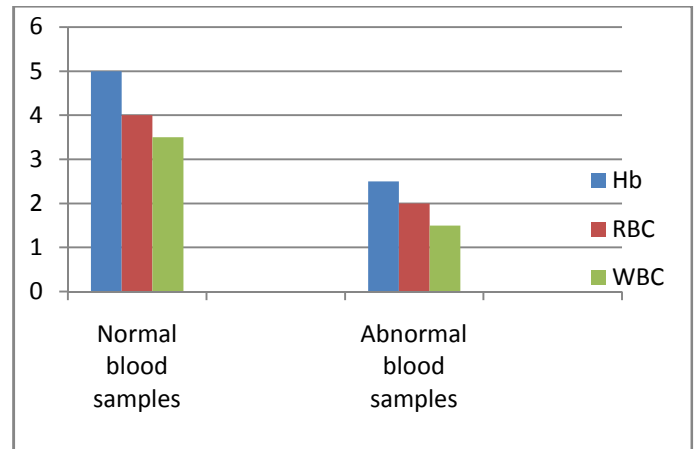


Figure 2. Show the comparison between normal and abnormal blood samples

From these results we find that the composition of menstrual blood show great variation and the content of blood like hemoglobin, RBC, WBC are present in fewer amounts in menstrual blood than the normal blood. On the basis of preliminary study we find that menstrual blood has less composition than normal blood samples that give a good parameter for differentiate the menstrual blood from normal blood samples.

Blood grouping

Menstrual blood gives positive results in determination of blood group by using antisera A, B and H.



Figure 3. Show the B positive blood group with menstrual blood

Microscopic analysis: Microscopically study of blood is very helpful in identify the type of blood from the crime scene and in this we observed that in menstrual blood number of cells are very low but dead cells and debris are highly present (shown in fig: 5). But in the normal blood samples RBC, WBC and other cells are present in highly amount (shown in fig: 4).

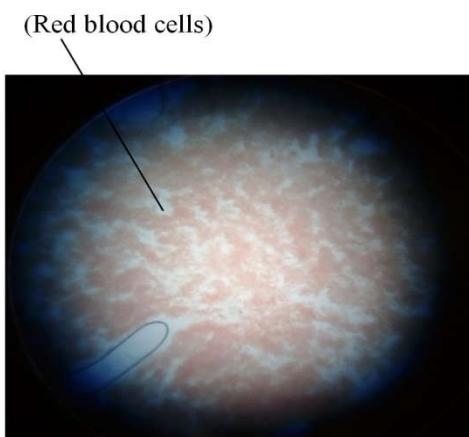


Figure 4. Show the Red blood cells Present in normal Blood sample

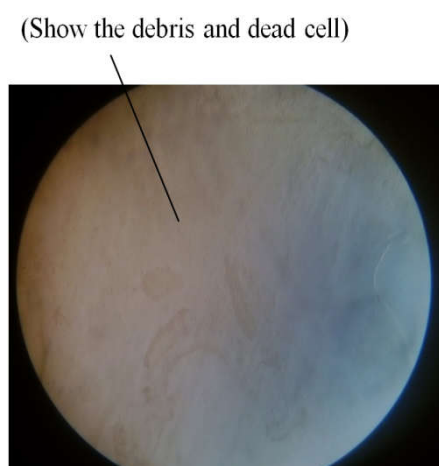


Figure 6. Show the negative results with menstrual blood

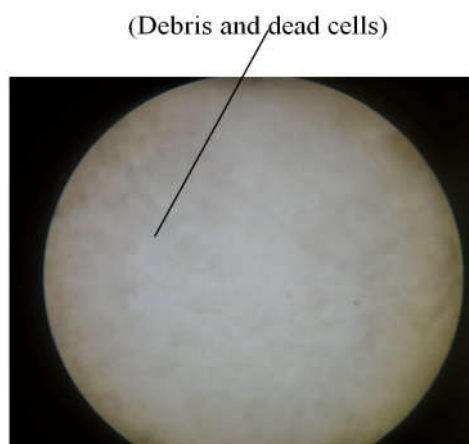


Figure 5. Show the debris and Dead cells Present in menstrual Blood

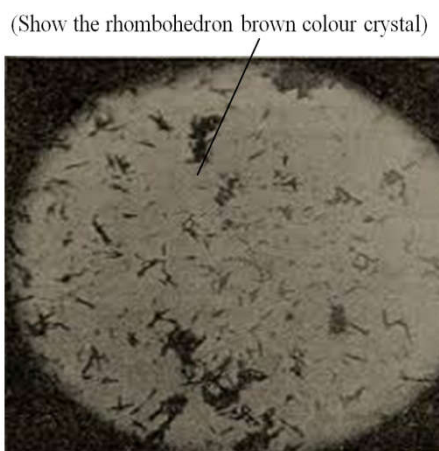


Figure 7. Show the positive results with normal blood

Which is very helpful in identify and comparison of normal and abnormal (menstrual) blood samples. In 1969 author was used electron microscope analysis of young and old Red blood cells stained with colloidal iron for surface charge evaluation. In this study electron micrographs of thin section of young cells showed uniform and dense deposition of positive iron particles in normal blood sample (Marikovsky and Danon, 1969).

Forensic analysis: Teichmann test show a negative results with menstrual blood but give a positive results with normal blood samples because hemoglobin level in menstrual blood is very low. In the case of normal blood sample hemoglobin level is very high so that hemoglobin has been react with teichmann reagent and gives positive results (fig.7). But hemoglobin is present in less quantity so hemoglobin not reacts properly with teichmann reagent so that it gives a negative result with menstrual blood

Conclusion

It is concluded that menstrual blood give a negative results with confirmatory test (teichmann test) of blood and the level of RBC and WBC is less in menstrual blood. It is very much useful for the identification of crime scene due to presence of debris and dead cell in the menstrual blood and the level of hemoglobin is very much less as compare to normal blood sample. By all these parameters it is a key step for the analysis of crime scene evaluation.

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