# Comparison of effects of cigarette and hookah smoking on human erythrogram: A Case control study

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Abstract—Smoking is a serious public health problem that leads to death thousands of people every year, being a variant responsible for many changes in laboratory tests. Thus, the aim of this study was to compare erythrogram of cigarette smokers and hookah smokers. Forty-five subjects participated in this study: cigarette smokers (15), hookah smokers (15) and non-smokers (15). Between October and December 2017, after the volunteers answered a questionnaire about smoking habits, peripheral blood samples were collected and analyzed for red blood cells (RBC) count, hemoglobin (Hb) content and determination of hematocrit (HCT) and hematimetric indices by using manual methods. Analyses were completed in 2018. Regarding the frequency of use, cigarettes are consumed in great amount every day, unlike the hookah, which is usually used only on weekends. However, when compared to control group, both cigarette and hookah smokers showed a significant increase in HCT, Hb and RBC values, but non-significant differences in hematimetric indices (p < 0.05). Cigarette and hookah smokers showed a similar and increased erythrogram profile. Although hookah has not been shown to be more harmful than cigarettes, variables such as the frequency and duration of hookah sessions can be directly related to the severity and intensity of changes, demonstrating evidence that it could be more dangerous to health than cigarettes. Therefore, more detailed studies on hookah should be conducted.

Keywords: Erythrogram, Cigarette, Hookah, Smoking.

# I. Introduction

Drugs are chemical substances that can modify all body functions, being divided into licit or illicit. Tobacco is a licit drug considered by the WHO to be a serious public health problem and the leading cause of avoidable death worldwide, since addiction and its degrees of intensity have complex mechanisms, making it difficult to quit smoking and generating high costs for the Unified Health System (SUS) with tobacco-related diseases. Tobacco smoke contains more than 4,720 toxic substances, such as nicotine, the active ingredient responsible for addiction; and carbon monoxide (CO), a toxic and carcinogenic gas that has a higher affinity for hemoglobin than oxygen. These substances can be found in any form of tobacco use, such as in cigarettes and hookah, harming both active and passive smokers exposed to these toxic substances.<sup>1-4</sup>

Nicotine acts in the brain through cholinergic receptors and its continuous use causes biological and physiological changes in the central nervous system. Thus, the brain of dependent people works differently from non-dependent.<sup>5</sup> Meanwhile, when tobacco smoke is inhaled, CO binds with hemoglobin to produce carboxyhemoglobin (COHb), preventing the body tissues to receive a continuous and appropriate oxygen delivery and increasing erythropoiesis, which leads to changes in some hematological parameters that modify the results of laboratory tests, in addition to increasing the risk of cardiovascular and respiratory diseases, several kinds of cancer and other tobacco-related diseases when there is prolonged exposure to smoke.<sup>6,7</sup>

Because of the public policies increased in recent years related to cigarette smoking, people end up looking for other forms of tobacco consumption, such as the use of hookah, due to the belief of being less harmful than cigarettes. However, some studies have already shown that the percentage of CO in hookah smoke is higher than that found in cigarettes, as well as many other toxic substances, becoming much more harmful to health. 4

In order to discourage this popular belief among youth and adults, some questions arise: what hematological changes can be observed in cigarette smokers? Would they be present in hookah smokers as well? Thus, this study aimed to compare the erythrograms of cigarette smokers and hookah smokers with matched control nonsmokers.

## II. METHODOLOGY

This case control type of observational study was done at Department of Biomedicine, Faculdade Fasipe, Sinop, MT, Brazil in year 2017-18.

This study was approved by the Research Ethics Committee of the CEI – Centro Educacional Integrado and an informed consent form (ICF) was obtained from all participants, according to the Resolution no. 466/12 of the National Health Council (CNS, acronym in Portuguese). The study was conducted in accordance with the Helsinki Declaration as revised in 2008.

#### 2.1 Study Population

This study was attended by 45 volunteers (age  $\geq$  18 years) from a college in the city of Sinop/MT, Brazil, randomly selected after responding to a questionnaire about their smoking habits and health condition. They were 15 in each group i.e. cigarette smokers, 15 hukkah smokers and 15 non-smokers. For smokers, smoking for more than 1 year was considered as inclusion criteria. Parameters such as the presence of anemia or other chronic pathologies were used as exclusion criteria. The individuals considered eligible to participate in the study were: non-smokers (9 female and 6 male), cigarette smokers (6 female and 9 male) and hookah smokers (7 female and 8 male).

The blood samples were collected and analyzed at FASICLIN, School Laboratory of FASIPE - Faculty of Sinop, between October and December 2017. Study was completed in year 2018.

#### 2.2 Measures

Peripheral blood samples for hematological analyses were collected by venipuncture and placed into EDTA containing tubes.

The following parameters were manually determined: red blood cells (RBC), hematocrit (HCT), hemoglobin (Hb), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC).

RBC count was done by Neubauer chamber and the cells were counted using a microscope at x40 objective lens. Microhematocrit method was used to determine the percentage of HCT by using capillary tubes in a microhematocrit centrifuge (3.500rpm/min) for 5 minutes, which was measured using a microhematocrit reader. Hb concentration was measured by the cyanmethemoglobin method with drabkin's reagent and the hematimetric indices were calculated from RBC count, Hb and HCT concentration using the following formulas<sup>9, 10</sup>: MCV = (PCV  $\div$  RBC) x 10 fL; MCH = (Hb  $\div$  RBC) x 10 pg; MCHC = (Hb  $\div$ PCV) x 100 g/dL.

# 2.3 Statistical Analysis

Data were complied and statistically analyzed by using GraphPad Prism version 7.0 (GraphPad Prism Software Inc., San Diego, CA, USA) and one-way variance analysis (ANOVA) followed by Tukey's test. Differences were considered significant when p < 0.05. The data are expressed as the mean  $\pm$  SEM for each group.

## III. RESULTS

In present study, most of the studied hematological parameters were affected with smoking whether of cigarette or of hookah except MCV (fL). (Table 1)

Table 1 Comparison of Hematological Parameters among Control Group, Cigarette and Hookah Smokers

Variables	Control	Cigarette	Hookah	P. Value LS
HCT(%) (Mean±SD)	$40.6 \pm 0.8$	46.5 ± 1.2*	45.8 ± 1.1*	<0.001 S
Hb(mg/dL) (Mean±SD)	$13.5 \pm 0.2$	15.5 ± 0.4*	$15.2 \pm 0.3$	<0.001 S
RBC(x106 /mm³) (Mean±SD)	$4.507 \pm 0.09$	5.187 ± 0.1*	5.067 ± 0.1*	<0.001 S
MCV(fL) (Mean±SD)	89.6 ± 0.4	$89.4 \pm 0.9$	$89.4 \pm 0.4$	0.592 NS
MCH(pg) (Mean±SD)	$29.8 \pm 0.2$	$29.4 \pm 0.3$	$29.6 \pm 0.1$	<0.001 S
MCHC (mg/dL) (Mean±SD)	$33 \pm 0$	$33.2 \pm 0.2$	$33 \pm 0$	<0.001 S

Mean HCT of control group was 40.6% which was significantly (<0.05) increase in cigarette as well as in hookah smoker group. When it was compared between cigarette and hookah group it was found not significantly (>0.05) increased in cigarette group than hookah group i.e. 46.5% v/s45.8%. (Table 1 & 2)

Mean Hemoglobine (Hb) of control group was 13.5 mg/dl which was significantly (<0.05) increase in cigarette as well as in hookah smoker group. When it was compared between cigarette and hookah group it was found significantly (<0.05) more increased in cigarette group than hookah group i.e. 15.5 v/s 15.2 %. (Table 1 & 2)

Mean RBC of control group was  $4.507 \times 106 \text{ /mm}^3$  which was significantly (<0.05) increase in cigarette as well as in hookah smoker group. When it was compared between cigarette and hookah group it was found significantly (<0.05) more increased in cigarette group than hookah group i.e.  $5.187 \times 106 \text{ /mm}^3 \times 106 \times 1000 \times 1$ 

Mean MCV (fL) of control group was 89.6 which was not significantly (>0.05) increase in cigarette as well as in hookah smoker group. When it was compared between cigarette and hookah group it was found not more of less equal i.e. 89.4 in both group with SD 0.9 and 0.4 respectively (>0.05). (Table 1 & 2)

Mean MCH of control group was 29.8 pg which was significantly (<0.05) increase in cigarette as well as in hookah smoker group. When it was compared between cigarette and hookah group it was found significantly (<0.05) more increased in cigarette group than hookah group i.e. 29.4 v/s 29.6 pg.(Table 1 & 2)

Mean MCHC of control group was 33 mg/dl which was significantly (<0.05) increase in cigarette but not in hookah smoker group. When it was compared between cigarette and hookah group it was found significantly (<0.05) more decrease in cigarette group than hookah group i.e. 33.2 v/s 33mg/dl.(Table 1 & 2)

Table 2 Comparison of Hematological Parameters in all three groups by Post hoc Tukey Test

Variables	Control v/s Cigarette	Control v/s Hookah	Cigarette v/s Hookah
HCT(%)	<0.05 S	<0.05 S	>0.05 NS
Hb(mg/dL)	<0.05 S	<0.05 S	<0.05 S
RBC(x106 /mm <sup>3</sup> )	<0.05 S	<0.05 S	<0.05 S
MCV(fL)	>0.05 NS	>0.05 NS	>0.05 NS
MCH(pg)	<0.05 S	<0.05 S	<0.05 S
MCHC (mg/dL)	<0.05 S	>0.05 NS	<0.05 S

## IV. DISCUSSION

Smoking is considered one of the main public health problems and the results of this study also show that the use of cigarette and hookah has negative effects on HCT values, RBC count and Hb concentration among the study population. The main cause of tobacco-related diseases is related to its continuous and intense use, being aggravated according to the number of cigarettes smoked per day. Comparing the frequency of smoking among cigarette smokers and hookah smokers, it was observed that cigarettes are consumed in large quantity every day, unlike the hookah, which presented a higher frequency of use only on weekends. The consumption of hookah seems to be associated with social and cultural characteristics, involving the "hookah sessions" in a group, used as a form of interaction among the young, while the use of cigarettes presents more complex aspects, ranging from genetic factors to psychological problems that lead to a complicated smoke addiction difficult to overcome, justifying the divergence in smoking frequency of both groups. Even so, the results of the evaluated parameters of both groups were similar, showing that the hookah can be as dangerous as the cigarette, or even more, depending on the frequency of smoking.

Results showed that HCT values were significantly higher in both cigarette and hookah smokers when compared to control group. Studies comparing the effects of cigarette and hookah on different hematological parameters of rats and humans also showed a significant elevation of HCT due to tissue hypoxia caused by the CO concentration present in both forms of tobacco use. When O2 transport is reduced, the body needs to increase RBC production by increasing the secretion of erythropoietin, which is reflected in HCT values.<sup>6, 11, 14, 15, 16,</sup>

Smoking is considered one of the main causes of HCT elevation. However, since the negative effects of tobacco arise in the long term, routine laboratory tests may detect significant changes only after a longer period of time. In such cases, the severity of the condition should be taken into consideration. In this study, cigarette smokers presented a mean of 12.7 ( $\pm$  2.8) years in terms of smoking time, compared to a mean of only 3.1 ( $\pm$  0.4) years among hookah smokers, which have nonetheless already shown altered results in HCT analysis.

The results indicated that both cigarette and hookah smoking led to increased Hb concentration when compared to control group. Jain et al.<sup>15</sup> and Khalaf et al.<sup>16</sup> also reported that Hb concentrations were significantly greater in smokers when compared to non-smokers due to increased sizes or numbers of RBCs, corroborating with the increased HCT values among smokers. When COHb molecule is formed and the oxygen delivery to tissues is disrupted, increasing erythropoiesis process, Hb concentration is also influenced.

Thus, COHb can also be used to determine the levels of exposure to CO through hookah and cigarettes. Zahran et al.<sup>20</sup> related increased COHb concentrations after a hookah session of 10-40 minutes, whose

results were higher than 10-40 minutes smoking cigarettes. Thereby, it is believed that COHb concentrations may be higher among hookah smokers than among cigarette smokers, depending on the duration and frequency of the hookah sessions. So, since high levels of Hb are related to increased COHb, cigarette and hookah smokers in this study may have shown similar results due to the low frequency of smoking reported by most of the hookah smokers. Otherwise, this latter group could have presented higher values of Hb.

Also, results indicated that hookah and cigarette smoke led to increased RBC count when compared to control group, corroborating with studies carried out by Shah et al.<sup>21</sup> and Miri-Moghaddam et al.<sup>14</sup> HCT and Hb elevation are related to increased RBC count due to a higher blood concentration of COHb, as a form of compensatory mechanism of the organism, which can mimic a secondary polycythemia condition by increasing capillary permeability.<sup>11, 15, 22</sup>

This study did not show significant differences between the HCT, Hb and RBC of cigarette and hookah smokers. However, a group of researchers showed significantly higher results of these parameters in the groups exposed to hookah smoke for a certain period, increasing according to the time of exposure, demonstrating that it may contain higher concentrations of toxic substances than cigarette smoke, being more harmful to health. <sup>14, 23</sup>

Regarding the MCV, MCH and MCHC, the results showed no significant differences between the three groups. These findings were similar to observations made by Jain et al.15 and Asif et al.25 However, according to some authors, smokers usually present higher MCV and MCH than nonsmokers, although their pathogenesis are not clearly understood. Besides the compensatory mechanism of the organism against tissue hypoxia, it is believed that the acetaldehyde toxicity on RBCs and the functional deficiency of vitamin B12 by cyanide action are also related to the increase in MCV, being reflected in MCH values, since the mean corpuscular volume can affect the hemoglobin content. 11, 24

Blood rheology, or study of the flow properties of its components, is influenced by the viscosity and deformity of RBCs. The higher the HCT or RBC, the greater the blood viscosity. However, the deformability depends on the cytoplasmic viscosity of these cells, which is mainly controlled by CHCM. The higher the CHCM, the less erythrocyte deformability. The same applies to the increase in VCM. Since the properties studied in the blood rheology play an important role in the development of occlusive arterial diseases, it is important to study the effects that smoking may have on it.<sup>25</sup>

#### V. CONCLUSION

This present study concluded that there was increased values of HCT, Hb and RBC in both cigarette and hookah smokers. Surprisingly hookah was found more harmful habit than cigarettes. It is necessary to consider some variables, such as the frequency and duration of hookah sessions, which show evidence that it could be more harmful to health, since it is inferred that the intensity of the changes is proportional to their consumption. More detailed studies are required to understand the characteristics of hookah.

#### **CONFLICT OF INTEREST**

None declared till now.

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