Sudan University of Science and Technology

College of Graduate Studies

Effect of Cannabis (Bango) on Complete Blood Cell Count among Sudanese Addicts

تأثير القنب (بنقو) على تعداد خلايا الدم الكامله بين المدمنين السودانيين

A dissertation Submitted in Partial Fulfillment For The Requirement of M.Sc. Degree in Medical Laboratory Science – Hematology and Immunohematology

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الآيـــــــــة

قَالَ تَعَالَىٰ:

﴿ قَالُواْ سُبۡحَٰنَكَ لَا عِلۡمَ لَنَآ إِلَّا مَا عَلَّمۡتَنَآۖ إِنَّكَ أَنتَ ٱلۡعَلِيمُ ٱلۡحَكِيمُ ٣٢ ﴾

صدق الله العظيم

سورة البقرة الاية "32"

Dedication

Every challenging work needs self-efforts as well as
Guidance of those who were very close to our heart
To my parents
Thanks for your great support and continuous care

To my brothers, and my sisters
To my friends and teachers
To all whom I love. . .

Acknowledgments

First of all I thank Allah, for all the physical and mental strength bestowed to me. I would like to express my sincere gratitude to my supervisor: Dr Selma E.Abdalla for his guidance, feedback and support throughout the study. Also special thanks to all members of Sudan university of science and technology, Collage of graduate studies especially to staff members of Hematology, I am really do not find the words the students, who helped me acquire new laboratory skills and were always available for assistance and advice.

My great thanks to laboratory staff at Al-Hayat Health Care Center Finally, my source of thanks to all participates in this study, for their time, patience, and dedication to research. Without you, none of this would be possible.

Abstract

This is a case control study, aimed to investigate the effects of cannabis and other substances on blood cells count, of Sudanese addicted in Khartoum state. The study conducted in al-Hayat health care center, during the period from September to October, 2018. Forty cannabis abuser and twenty healthy volunteers, were enrolled in this study after a written consent had been obtained from them, the data were collected using a questionnaire and laboratory measurement. Venous blood of 2.5ml was collected in K_3EDTA containers and CBC parameter was determined using automated hematological analyzer (sysmex). The data analyzed by SPSS version 19.

The distribution of cannabis abusers according to age recorded the highest frequency (13) by the age 27-29 years old and the lowest frequency (1) by the age 36-38 years old. The secondary educational qualification recorded the highest frequency (26), and (3) for technical qualification. Duration of (3-5) years of cannabis addicts recorded the highest frequency (23), and (9-11) years recorded the lowest frequency (4).

The distribution of other substances addicted with cannabis indicated that cigarette recorded the highest frequency (14), and tobacco recorded the lowest (1). The abuser jobs indicated that casual jobs recorded highest frequency (23), and permanent job. Hemoglobin concentration were significantly higher in cannabis abuser compared with control group (14.0±2.8 vs 12.6±2.6). Platelet count was significantly lower in cannabis abuser compared with control (260.0±69.29 vs 327.9±70.14).

Neutrophils count was significantly lower in Cannabis abuser group compared to the control group (35.09±9.3 vs 40.3±12.6). WBCs, RBCs, HCT, and RDW-SD values were not influenced significantly by cannabis addiction. Our results point out the risk of increased bacterial infection due to a long-term use of Cannabis. In addition, measures to control the spread of Cannabis abuse should be implemented.

خلاصة

هذه دراسة الحاله والتحكم، تهدف الي تقييم تأثير إدمان القنب (بنقو) ومواد مخدره أخرى على تعداد خلايا الدم، للمدمنين السودانيين داخل ولاية الخرطوم، أجريت هذه الدراسة في مركز صحي حياة، خلال الفترة من سبتمبر الى أكتوبر2018. أجريت هذه الدراسة في 40 مدمن على البنقو و 20 متبرع غير مدمن (صحي)، بعد الحصول على الموافقة ، تم جمع المعلومات بواسطة الإستبيان والفحوصات المعملية، تم جمع 2.5مل من دم الوريد، داخل حاويات EDTA، تم تحديد مقياس الدم بإستخدام جهاز تحليل الدم الأتوماتيكي (سيسميكس)، ثم تحليل البيانات عن طريق SPSS إصدار19.

توزيع مدمنين البنقو طبقًا الى الأعمار سجل أعلى تكرار (3) للأعمار بين (27-29)عام، وأدنى تكرار(1) للأعمار بين (36-38)عام. المؤهل التعليمي الثانوي سجل أعلى تكرار للمدمنين (26) مقارنة بأقل تكرار للمؤهل التقني (3). المدة (3-5)سنة من إدمان البنقو سجلت أعلى تكرار(23)، مقارنة بأدنى تكرار(4) للفترة بين (9-11). إنتشار تعاطي البنقو مع مواد أخرى أشارت الي تدخين السجائر بأعلى معدل (14) مع البنقو، مقارنة بتكرار التمباك (1).

الوظائف المؤقته بين المدمنين سجلت أعلى تكرار (23) مقارنة بالوظائف الدائمة (17) سجلت اقل تكرار. تركيز خضاب الدم معنويا أعلى في المدمنين مقارنة بغير المدمنين. إنخفض عدد الصفائح الدموية معنويا في المدمنين مقارنة بمجموعة المتحكمين. نقصان الخلايا الحبيبية معنويا في المدمنين مقارنة بغير المدمنين. لم تتأثر قيم كل من RBCs, WBCs, HCT, SD-RDW معنويا بإدمان البنقو.

أشارت نتائجنا الى زيادة خطر الإصابه البكتيريه الناجمة عن إستخدام البنقو، بالإضافه الى ذلك ينبغي تنفيذ تدابير للسيطرة على إنتشار البنقو.

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Chapter One

Introduction & Literature review

CHAPTER ONE

INTRODUCTION AND LITERATURE REVIEW

1.1 Introduction:

Cannabis sativa has been very popular all over the world, it is used for diverse purposes which range from medicinal use, and recreation, to severe forms of dependency, synthetic cannabinoids represent an increasingly popular trend, and acute intoxication is widely seen in emergency rooms and intensive care unit, as they are inexpensive and easily accessible. Use of synthetic cannabinoids has psychiatric, medical, and social consequence. They are found in powder form or loose leaf form. Powder form may be dissolved and sprayed onto a dried herbal substrate, and they are typically smoked in cigarette papers or cannabis pipes (Spaderna, et al., 2013).

The physical harm caused by cannabis is less well-known. In adults, chronic bronchitis, lung cancer, myocardial infarction, hepatotoxicity, decreased sperm count and motility, gynaecomastia in males, suppression of ovulation among females, low birth weight and delayed visual system development among the newborns of cannabis using females (Rizwana, 2015).

Plant derived cannabinoids, include delta-9-tetrahydrocannabinol (THC), the primary psychoactive component of cannabis. Cannabinoids mediate their effects through binding specific receptors which are members of the G protein coupled receptor superfamily. Two cannabinoid receptors have been identified: Cannabinoid-1 receptor (CB1) and cannabinoid-2 receptor (CB2). CB1 are expressed primarily in the central nervous system (CNS), and are responsible for the

psychoactive effects of cannabinoids by modulating neurotransmitter release (Brady, et al.,2009). In contrast, CB2 are localized primarily in immune cells such as lymphocytes, macrophages, and neutrophils and are responsible for immunodulatory effects of cannabinoids (Lehmann, et al., 2011, and Sardinha, et al., 2013).

Therapeutic effects of marijuana and THC, which include lowering of internal eye pressure in persons suffering from glaucoma and alleviating nausea and vomiting, caused by chemotherapeutic drugs used to treat cancer patients, and AIDs. Most continuous use of the plant is for its psychoactive or narcotic properties (Baczynsky and Zimmerman, 1983).

1.2 Literature review:

The blood is circulated around the body through blood vessels by the pumping action of the heart. And venous blood carries carbon dioxide, a waste product of metabolism produced by cells, from the tissues to the lungs to be exhaled. Blood perform many important functions within the body, including: supply of oxygen to tissue (bound to hemoglobin, which is carried in red cells), supply of nutrients such as glucose, amino acids, and fatty acids (dissolved in the blood or bound to plasma proteins), removal of waste such as carbon dioxide, urea, and lactic acid, immunological function, coagulation, messenger function, regulation of core body temperature, blood accounts for 7% of the human body weight, (Alberts, 2012).

Blood composed of plasma and several kinds of cells. These blood cells (which are also called corpuscular) consist of erythrocytes (red blood cells, RBcs), leukocyte (white blood cells, WBCs), and thrombocytes (platelets). By volume, the red blood cells constitute about 45% of whole blood, the plasma about 54.3%, and white cells about 0.7%, (Alertn, 2012).

1.2.1 Constituents of blood:

1.2.1.1 Red blood cells (RBCs):

Erythrocyte are produced in the bone marrow with the help of metals (iron, cobalt, manganese) vitamins (B12, B6, C, E, folate, riboflavin, pantothenic acid, thiamine), and amino acids. Is regulated by erythropoietin, thyroid hormone, and androgens. The mature red blood cell carries oxygen attached to the iron in hemoglobin, and a nucleated structure with no capacity to synthesize protein, yet it is capable of limited metabolism, which enables to survive 120 days. (Lokwani ,

2013). The author added that the number of red blood cells is determined by age, sex, altitude, exercise, diet, pollution, drug use, tobacco/nicotine use, kidney function, health disease status, etc. The clinical importance of the test is that it is a measure of the oxygen-carrying capacity of the blood.

Normal red blood cells: 4-6 million/mm.

1.2.1.2 Hemoglobin (Hb):

The main function of red cells is to carry oxygen to the tissue and return carbon dioxide from the tissue to lungs. Each molecule of normal adult hemoglobin A (HbA), consist of four polypeptide chains (2alph and 2beta), hemoglobin synthesis occurs largely in the mitochondria by a serious of biochemical reactions.

Hemoglobin normal range for male: 13.3-16.2 g\l, (Hoffbrand, 2011).

1.2.1.3 Hematocrit- packed cell volume (PCV):

The hematocrit (Hct) or packed cell volume (PCV), is the ratio of the volume of erythrocytes to that of whole blood and is reported as percentage.This is the earliest RBC parameter identified and is used for red cell disorders differentiation. High hematocrit due to: dehydration, kidney disease, low oxygen level in the blood, smoking, etc. Low hematocrit due to: blood loss, bone marrow failure, leukemia, etc, hematocrit normal range for male: 38.8-46.4%, (Lokwani, 2013).

1.2.1.4 Mean corpuscular volume (MCV):

MCV is average volume of the red blood cell. Is a useful red cell index that is used in classification of anemia's, the MCV is measured in femtoliters(fl, or 10^{-15} L), using automated methods this value is derived by dividing the summation of the red cell volumes by the erythrocyte count. MCV= hematocrit (L/L) x 1000 / red cell count(x10^{12} /L).

MCV normal range for men & women: 79-98fl, (Wintrob's, 1999).

1.2.1.5 Mean corpuscular hemoglobin (MCH):

MCH is a measure of the hemoglobin content per red cell. It may be calculated manually or by automated method using the following formula: MCH= hemoglobin (g/l) / red cell count $x10^{12}$ /L. MCH is expressed in pictograms (pg or 10^{-12} g), MCH reflects the mass of hemoglobin, MCH measurement may be falsely elevated by hyperlipidemia or leukocytosis, because increase plasma turbidity may erroneously elevate the hemoglobin measurement.

Normal range of MCH: 26.7-31.9pg\cell, (Wintrob's, 1999).

1.2.1.6 Mean corpuscular hemoglobin concentration (MCHC):

The average concentration of hemoglobin per red cell may be calculated by following formula:

MCHC= hemoglobin(g/dl) / hematocrit (L/L). The MCHC is expressed in grams of hemoglobin per deciliter of packed red blood cells. This measures the concentration of hemoglobin , the determination of MCHC is affected by conditions that affect either hematocrit (plasma trapping or presence of abnormal red cells) or hemoglobin (hyperlipidemia, leukocytosis).

MCHC normal range: 32-36g\dl. (Wintrob's, 1999).

1.2.1.7 Red cell distribution width (RDW):

Modern analyzers also record the red cell distribution width (cell volume distribution). In normal erythrocyte morphology, this correlates with price-jones curve for the cell diameter distribution. (Harald, et al, 2002).

RDW-SD:

The determination of the RDW-SD is an actual measurement of the width of the erythrocyte distribution curve; these measurements are performed at a relative height of 20% above the baseline. The wider the

curve is spread by erythrocytes of different sizes, the higher the RDW-SD value will be. Reference value: 35-45fl, (Lokwani , 2013).

1.2.1.8 White blood cells (WBCs):

The enumeration of leukocytes, usually called the white blood cell count, is one of the most frequently obtained laboratory values in medicine, because it is extremely informative as to the state of health of the host. The blood contains three types of white blood cells : the polymorphnuclear leukocyte, the monocyte, and the lymphocytes, these leukocyte arise feom pluripotent stem cells in the bone marrow, and differentiate into those forms that are easily distinguished and different numbers of which circulate in the blood, (Babior and stossel, 1994).

1.2.1.8.1 Granulocytes (polymorph nuclear leukocytes):

Leukocytes characterized by the presence of differently staining granules in their cytoplasm when viewed under light microscope. These granules are membrane-bound enzymes which primarily act in the digestion of endocytosed particles. There are three types of granulocytes: neutrophils, basophils, and eosinophil's, which are named according to their staining properties, (Lokwani , 2013).

A. Neutrophil (polymorph nuclear):

Have no preference for acidic or basic stains, diameter; 12-13um, cytoplasm; pink, nucleus; dark purple to blue with dense chromatin;2-3 lobules. Causes of increased neutrophil (neutrophilia): physiologic increase, tissue injury and inflammation, neoplasms, acute bacterial infection, etc. Causes of decrease neutrophil (neutropenia): decrease or ineffective production as aplastic anemia, and increase removal from circulation.

Percentage of total white blood cells: 40-80%, (Lokwani, 2013).

B. Eosinophil:

These cells are similar to neutrophils, except that the cytoplasmic granules are coarser and more deeply red staining and there are rarely more than three nuclear lobes, they enter inflammatory exudates and have a secial role in allergic responses, defense against parasites and removal of fibrin formed during inflammation, Percentage of total white blood cells: 1-4%, (Hoffbrand, , 2011).

C. Basophils:

Basophil undergo a maturation process similar to that described for neutrophil. Matyre basophil is about 10-15um in size and has segmented nucleus. They have large purple black coloured granules unevenly distributed throughout the cytoplasm completely obscuring both the cytoplasm and nucleous, (Shah and Bajaj, 2014).

Percentage of total white blood cells: 0.5-2%.

1.2.1.8.2 A granulocytes (mononuclear leukocytes):

Leukocyte characterized by the apparent absence of granules in their cytoplasm. Although the name implies a lack of granules these cells do contain non-specific azurophilic granules, which are lysosomes. The cells include lymphocytes, monocytes, and macrophage, (Lokwani, 2013).

A. Monocytes:

Monocytes are frequently noted for having vacuoles throughout their life cycle and under normal conditions. Except for the monocyte, vacuolization of the cytoplasm is commonly seen in older cells and in normal condition, sever bacterial infections, viral infections, and malignancies, may produce a remarkable number of vacuoles in various leukocyte types, (Mary, 2012).

B. Lymphocytes:

There are two types of lymphocytes, small lymphocyte, and large lymphocyte.

1-Small lymphocytes: about 90% of the circulating lymphocytes are small cells (9-12um in diameter), with a purplish nucleus, a thin rim of cytoplasm devoid of granules surrounds it. About two-thirds of these cells are T lymphocytes and most of the remainder is B-lymphocytes, but these two types are indistinguishable on routine smears.

2-Large lymphocytes: about 10% of the circulating lymphocytes are larger (12-16um in diameter) than the small lymphocytes, have a more abundant cytoplasm, possess a less condensed nuclear chromatin. Percentage of total white blood cells: 20-40%. (Lokwani, 2013).

1.2.1.9 Thrombocyte (platelets):

Platelets production follows formation of microviscle to form platelet demarcation membranes. The time interval from differentiation of the human stem cell to the production of platelets averages at about 10 days. Thrombopoietin is the major regular of platelet production and is constitutively produced by the liver and kidneys. (Hoffbrand, et al., 2001).

1.2.1.10 Mean platelet volume (MPV):

Is a measurement that describes the average size of platelet in the blood; this parameter provides an indicator as to whether the bone marrow is manufacturing platelets normally or there is some kind of production pressure from periphery like excessive destruction. Have inverse relationship with platelet number, MPV normal range: 7.4-10.4fl, (Lokwani , 2013).

1.2.2 Definition of addiction:

Is a brain disorder characterized by compulsive engagement in rewarding stimuli despite adverse consequences. Despite the involvement of a number of psychosocial factors, a biological process-one which is induced by repeated exposure to an addictive stimulus-is the core pathology that drives the development and maintenance of an addiction, (Nestler and Russo, 2013).

1.2.2.1 Addictive Disorder (or Addiction):

The essential feature of this disorder is a cluster of cognitive, behavioral, and physiologic symptoms that indicate that the person has impaired control of psychoactive substance use and continues use of the substance despite adverse consequences, (Smith, 2012).

The effects include mood disorders euphoria, anxiety, psychosis, cognitive and neurological function disorders (dizziness, seizures, tremor, and ataxia). Cardiovascular effects include tachycardia, bradycardia, ECG change and arrhythmias. Synthetic cannabinoids also induce dry mouth, redded conjunctiva, pupillary changes including miosis and mydriasis, blurry vision and light sensitivity. Hyperglycemia, hypokalemia, elevated creatinine, acidosis, elevated phosphokinase and elevated white blood cell count are reported, (Spaderna, et al., 2013. And; Altinisik, et al., 2015).

1.2.2.2 Cannabinoid addiction:

Cannabis, more commonly called marijuana, is a tobacco-like greenish or brownish material consisting of the dried flowers, fruiting tops and leaves of the cannabis plant. Hashish or cannabis resin is the dried brown or black resinous secretion of the flowering tops of the cannabis plant and can be further processed to produce hash oil, wax or shatter (NIDA, 2015). Also, there is preliminary evidence of higher risks of resins due to the higher concentrations of THC. Cannabis is usually

smoked as a cigarette (“joint”) or smoked as resins in a pipe or bong, also known as “dabbing” Vaporizers are also used to reduce the toxins inhaled from cannabis smoke.

Cannabis is reported to be the most abused drug in Sudan, followed by Diazepam. Other drugs abused include barbiturates, heroin and cocaine. (U.N, 1991). A plant called "Argemone Mexicana" from the papaveraceae family, which grows in different areas of the country (U.N. 1992) The overall prevalence of substance use is 31%. The all prevalence of tobacco, cannabis, alcohol, amphetamines, tranquilizers, inhalants, opiates, cocaine, and heroin use was 13.7%, 4.9%, 2.7%, 2.4%, 3.2%, 1%, 1.2%, 0.7%, and 0.5%, respectively(Osman et al, 2016).The authors also added that Curiosity (33.1%) was the main reason for initiation of substance use.

Most synthetic cannabinoids are not currently found using routine toxicology screening as they contain synthetic cannabinoids from different chemical classes. Can be measured in the serum using chromatographic methods with mass spectrometry but these test cannot be performed in many hospital and they time consuming an expensive (Scavnicar, et al., 2015).

1.3 Rationale:

Tetra hydro cannabinol (THC) is metabolized in the liver by microsomal hydroxylation and oxidation catalyzed by enzymes of cytochrome P450 (CYP) complex into an active compound, 11-hydroxyTHC (11-OH-THC) which is further metabolized into inactive forms. In type of overdosing active form is accumulating in liver and cause liver damage.

Also decrease of neutrophil count lead to increase of lymphocyte count. As a result, a greater risk for bacterial infection. The neutrophil to lymphocyte ratio has been widely used in identifying the degree of inflammation.

Many studies reported the effect of cannabis on Blood cells. The published data in Sudan is rare.

1.4 Objectives:

1.4.1 General objective:

-To assess the effect of Cannabis and other addicted substances on blood cells among Sudanese cannabis addicted in Khartoum State.

1.4.2 Specific objectives:

1- To compare the means of CBC parameter (RBCs, WBCs, Plt, Hb, Hct, RDW-SD, Neutrophils count, and Lymphocytes count) between cannabis addict and healthy people.

2- To determine the frequency of variables (age, educational qualify, nature of job, duration, addict with other substance), according to cannabis addicts.

3- To investigate the association of duration, age, addict with other substances and CBC parameter.

Chapter Two

Materials & Methodology

CHAPTER TWO

MATERIALS AND METHODOLOGY

2.1 Study design:

The case control study was conducted to estimate the effect of cannabis and other substances on blood cells count in Khartoum state.

2.2 Study area and duration:

The study was carried out at Al-Hayat Health Care Center, Khartoum through the period (September to October, 2018).

2.3 Study population:

The analytical study population was two groups: Sudanese addicted cannabis and other substances addict was considered as case group (N =40). Healthy Sudanese were consider as control group (N =20).

2.3.1 Inclusion criteria:

Sudanese Addicted cannabis.

2.3.2 Exclusion criteria:

- Addicts with history of anemia, thrombocytopenia & infections.
- Addicts not use cannabis.

2.4 Data collection:

Data were collected using a questionnaire, which was designed to collect and maintain all valuable information concerning each case examine,

2.5 Sample collection and processing:

Venous blood 2.5ml was collected from each participant in the

study. The sample collected under aseptic conditions and drawn into K3EDTA containers, and then samples were kept until the time of analysis.

2.6 Hematological analysis:

A complete blood count (CBC) and differential was performed on the blood sample using Sysmex KX-21N, an automated 3-part differential hematology analyzer (Sysmex Corporation Kobe, Japan) at the laboratory of Al-Hayat Health Care Center. Standardization, calibration of the instrument, and processing of the samples were done according to the manufacturer's instructions. The machine automatically dilutes whole-blood sample of 50 ml in the CBC/Differential mode, lyses and enumerates white blood cells (WBC), red blood cells (RBC), hemoglobin concentration (Hb), pack cells volume (PCV), platelets, lymphocytes, neutrophils and red blood cell indices (MCV, MCH & MCHC). It however does not count for eosinophil, monocytes and basophils counts.

2.7 Ethical consideration:

The study was approved by the Medical Laboratory college committee- SUST, Informed consent will be taken. Patients informed by the results. Personnel identification data will be kept secure.

2.8 Statistical analysis:

Data was analyses by using SPSS (version 19).Qualitative data was represented as frequency and percentage. Quantitative data was presented as mean ± SD. Correlation between variables was tested using Pearson's Chi square.

Chapter Three

Results

CHAPTER THREE

3. RESULTS

The some tables of results were presented in the appendix.

3.1 Demographic data:

The case and control study aimed to estimate the effect of cannabis and other substances on blood cells count. Sixty Sudanese subjects were enrolled in this study, 40 patients with cannabis addict and 20 healthy volunteers as control group. Figures (3.1) show the frequency of addict.

The distribution of the cannabis addict according to age were show in figure (3.2), the highest frequency (13) was recorded by the age group (27-29) years old, and the lowest frequency (1) by the group of (36-38) years. Nature of job among cannabis addiction, figure (3.3) show that casual job recorded highest frequency (23), and permanent job recorded lowest frequency (17).

The distribution of cannabis addicts according to the duration of addiction has been show in figure duration of (3-5) years recorded the highest frequency (23), and (9-11) years to the lowest frequency (4), figure (3.4).

The distribution of cannabis addicts according to their educational qualification, show that secondary educational qualification recorded highest frequency (26), and technical qualification recorded the lowest frequency (3), Table (3.5).

Distribution of cannabis addict with other substances, Table (3.8) indicates that cigarette recorded the highest frequency (14) for the other substances with cannabis addiction and tobacco recorded the lowest (1).

3.2. Hematological analysis:

The result of the study showed that two groups significantly differed on Hemoglobin, Platelet count, Neutrophils count, and lymphocytes count, while on the other hand RBCs, WBCs, Hct, and RDW-SD (P value > 0.05) showed non-significant differences between cannabis abusers and control group on blood cell count. Cannabis abuser showed (P <0.04) higher value of hemoglobin concentration (14.0±2.8), in comparison with control group (12.6±2.9).

Cannabis abuser showed lower value in platelet count (260± 69.29) in accord with control group (327.9±70.14), and (P value = 0.01). In the context of neutrophils the cannabis abusers showed lower value (35.09±9.3) in perspective with control group (40.3±12.6) and (P value = 0.02). The lymphocyte count in cannabis abusers (49.39±8.0) compared with control group (35.4±7.34), the RBCs in cannabis abusers (4.5 ±1.5) compared with control group (4.5±1.9), the WBCs in cannabis abuser (7.5±2.8), compared with control group (7.4±2.3), the hematocrit among cannabis abuser (38.9±9.2) compared with control (37.5 ± 8.0). RDW-SD among cannabis abuser (43.6± 6.3) compared with control group (42.6± 6.3). Table (3.3).

There was a significant positive correlation between hemoglobin, platelets, neutrophils count and duration (P=0.02) as shown in Table (3.4). There was a significant positive correlation between hemoglobin level and age (P=0.05). Show in Table (3.4). There was no correlation between hemoglobin, platelets, neutrophils count and addict with other substances (P= 0.59). Show in Table (3.4).

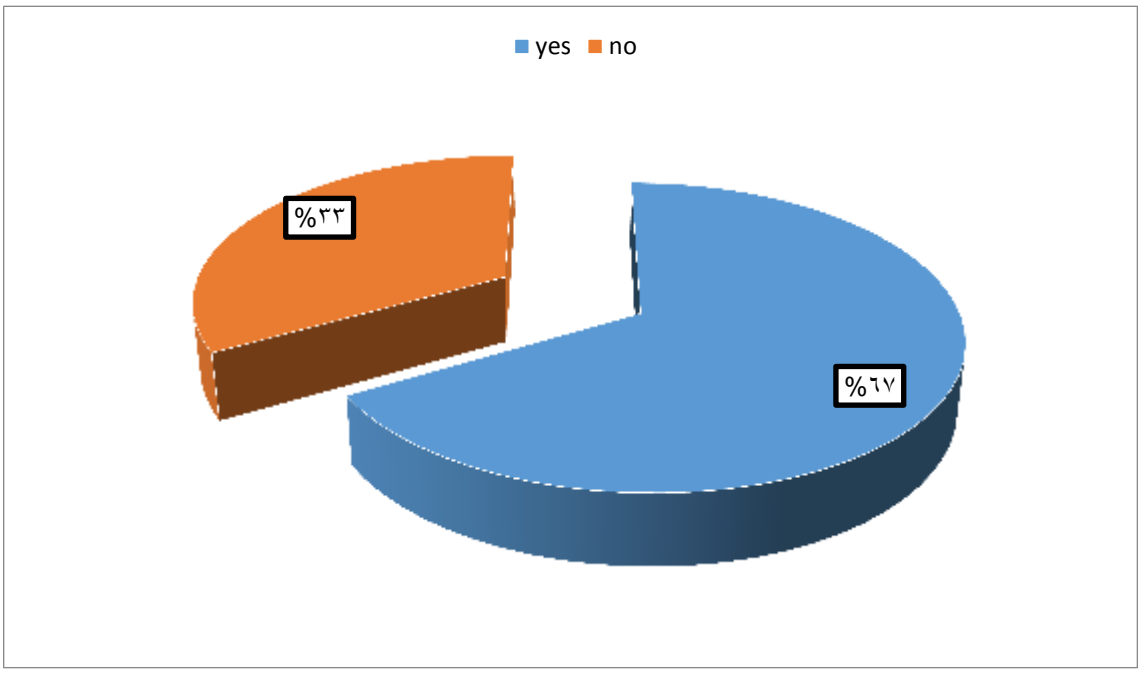

Figure**3.1** The distribution of subject according to addict

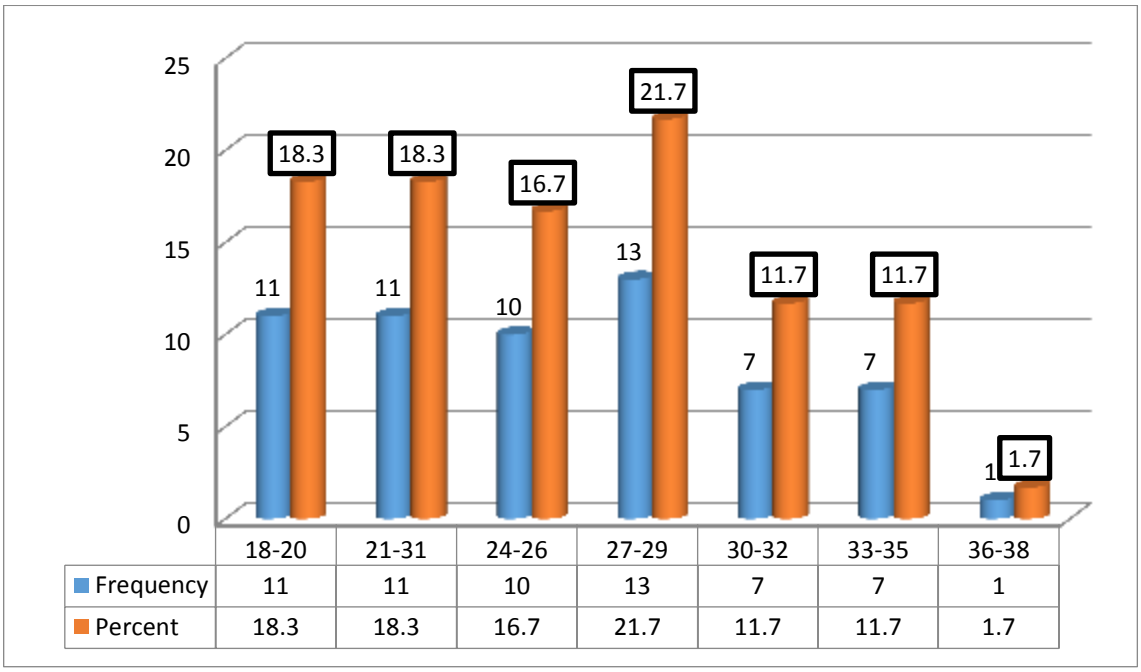

Figure3.2 The distribution of subject according to their age

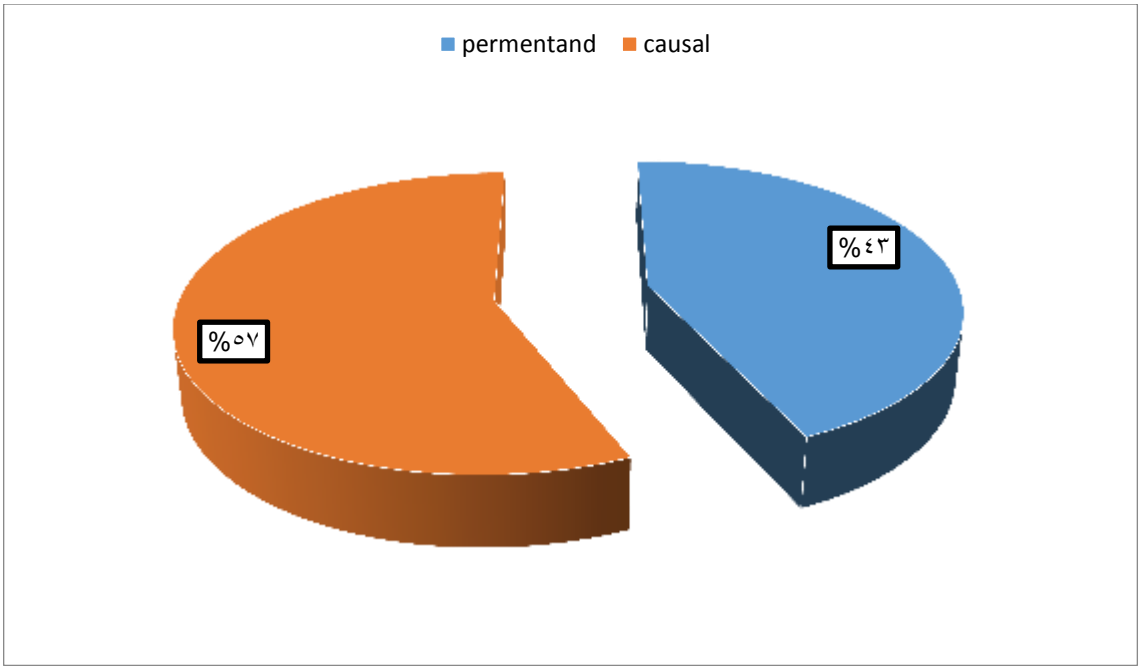

Figure3.3 The distribution of subject according to their nature of job

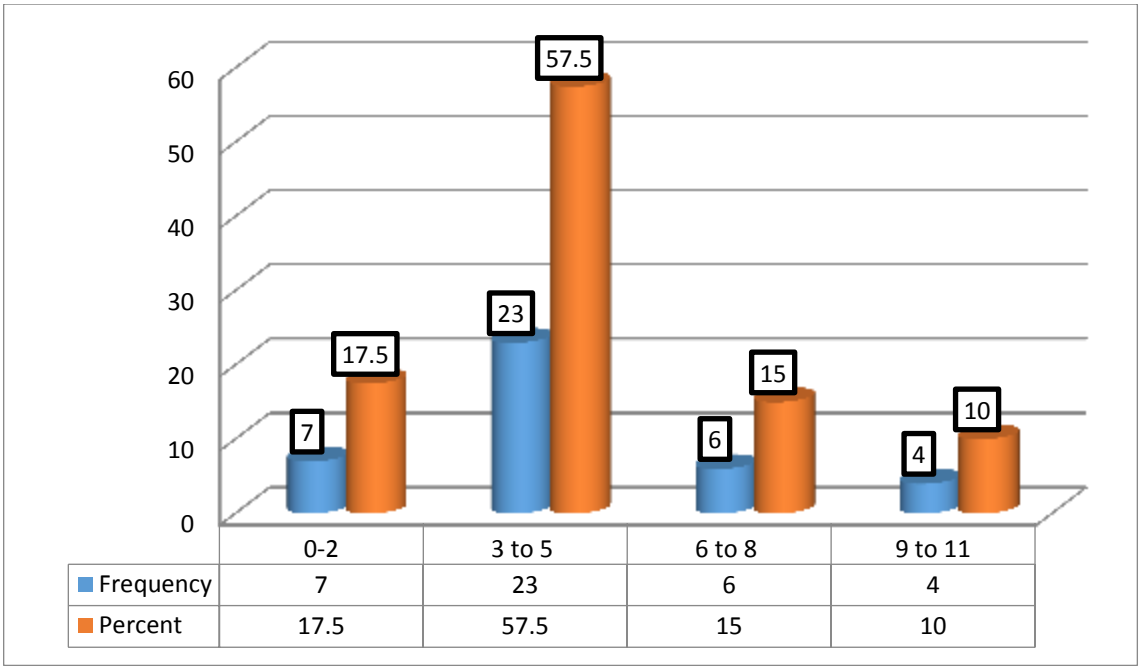

Figure4.4 Distribution of cannabis addict according to duration

Table 3.3 Comparison between Means of hemoglobin, platelets, neutrophils, lymphocytes counts in case and control group

Variables	Case (40) Mean ± SD	Control (20) Mean ± SD	P.Value
Hemoglobin	14.0 ± 2.8	12.6 ±2.9	0.04
Platelets	260± 69.29	327.9 ±70.14	0.01
Lymphocytes	49.30±8.00	35.4 ±7.34	0.01
Neutrophils	35.09 ±9.339	40.3 ±12.62	0.02
RBCs	4.5±1.5	4.5±1.9	0.93
WBCs	7.5±4.8	7.4±2.3	0.07
Hematocrit	38.9±9.2	37.5±8	0.17
RDW-SD	43.6±6.3	42.6±6.3	0.06

P value less than 0.05 are considered significant. SD: Standard Deviation

Table 3.4 The correlation (P value) of hemoglobin, platelets, neutrophils count with variables

Variable	R value	P. value
Age	**0.61**	**.05**
Addict with other substances	**0.04**	**0.592**
Duration	**0.35**	**.02**

Table 3.5 The distribution of subject according to their educational qualification

Education	Frequency	Percent
Basic	7	17.5
Secondary	26	65
Graduate	4	10
Technical	3	7.5
Total	40	100

Table 3.8 Distribution of cannabis addict with other substances

With other substance	Frequency	Percent
Tobacco	1	2.5
Cigarette	14	35
Hookah -Tobacco	2	
Tobacco -Cohol-Cigarette	3	5
Tobacco-Cigarette	7	7.5
Tobacco-Cigarette-Hookah-Cohol	6	10
Cigarette-Cohol	3	15
Cigarette- Hookah	4	25
Total	40	100

Chapter Four

Discussion, Conclusion and Recommendation

CHAPTER FOUR

Discussion, Conclusion, and Recommendations

4.1 Discussion

This study estimates the effect of cannabis and other substances on blood cell count. The distribution of age value on cannabis addict with highly among 27-29 years, followed by age of 21-26 years, which shows on Figures (3-2). This study agree with (Lead, et al, 2018), who reported that for youth cannabis use was associated with psychotic symptoms following age 22, with depressive symptoms from age 16-19 and following age 25. Also the study on the line of (Ali, 2017) who reported that the highest frequency (55) was reported by the age of (20-35) years old of smokers.

The current study shows an increase in value of hemoglobin concentration and numerous increase of hematocrits in cannabis addict , also, some previous study have reported effects on the blood including; increases in hemoglobin concentration , packed cell volume, and red blood cell counts, and these have been attributed to chronic exposure to smoke and carbon monoxide similar to that tobacco smokers, (Gabbay, et al., 2005), but some studies record no significant effect for cannabis on hemoglobin concentration in marijuana smokers (Oseni, et al., 2006), on other hand,(Karimi, et al., 2007) reported that chronic consumption of cannabis lead to increase Hb concentration of cannabis abusers in Khashmiri youth.

The red blood cell obtained in this study insignificantly in case group compared with control group, this confirms the study by (Beacofield, et al., 1973), who found no effect of pipe and cigar smoking on hematological values, and in this line This confirms the study by (El-shahat, 2011) who obtained a higher red blood cell count in the test group.

The total white cell count obtained in this study was marginally higher in the case group compared with control group, but not statistically significant ($P>0.05$). The variations observed were all in line with the reports of (Klein, et al., 2003; Oseni, et al., 2006; El-shahat, 2011); but

contrary to the finding by (Amna and Nabiala, 2011) who found no significant changes between smokers and non-smokers. The slightly Lower hemoglobin values obtained by addicted men groups compared to control group in their study could be due to poor nutrition of the smokers.

Possible complications arising from additional inflammatory agent, who might complicate the bronchitis experienced generally in smoker.

The current study shows low values of neutrophil in cannabis addict, Table (3.3). The present results agree with the findings of (Oseni and Togun, 2006). Some studies were observed the different effects of cannabinoids on peripheral leukocyte counts, and (Yahya and Wastone, 1987), they showed an increase in eosinophil counts, and peripheral blood lymphocytes. The reduction of the number of the neutrophils caused by cannabinoids was reported in previous studies by (Murikinati, et al., 2010) who suggested that the activation of cannabinoid 2receptor (CB2), reduced ischemic injury and this action involved the reduction of the number of neutrophils in the ischemic brain of experimental mice.

The significantly increased lymphocyte percent observed in this study agrees with the findings by (Amna and Nabiala, 2011; El-shahat, 2011) but is contrary to the low lymphocyte count observed by (Oseni, et al., 2006). For the most part, invivo and invitro studies suggest that marijuana (THC) is an immune modulator and has immunosuppressive effects on T and B lymphocytes (Bhargava, 1996), as well as natural killer (NK) cells (Kusher, et al., 1994; Klein, et al., 1998), macrophages (Baldwin, et al., 1997), and neutrophils (Murikinati, etal., 2010).

The current study show low values of platelet compare with control group, Table (3.3).The current result agree with the observations of (Aronow and Cassidy, 1975) who reported that cannabis causes an increase in carboxy hemoglobin, a resultant increase in myocardial

oxygen demand, decrease in oxygen supply as well as an induction of platelet. Other study demonstrate marijuana may increase factor vii activity, however, there are mixed results in terms of the effects of smoked marijuana on platelet function (Heiden, et al., 1980).

The cannabis and other substances user profiles are shown in Table 3.8, cigarettes were the most common form of addict with cannabis (35%, (n=14)), other substances were co-dependent.

The duration of cannabis use varied from months to eleven years, and (57.5 %,(n=23)) of the subjects had been using it for 3-5 years.

The subjects in this study were adult and educated, but their rate of secondary educational was significantly high (65%, (n=26)) show Table (3.5). Some of limitations of this study were as follow; a small sample size, co-dependance on other substances, and retrospective method of data collection.

4.2Conclusion:

Some haematological characteristics of cannabis addict differ significantly from non-addict. The result of this study reported lower platelets and neutrophils count in addicted cannabis people compared with healthy people, neutropenia makes an individual highly susceptible to infections.

Hemoglobin concentration significantly increases; there were statistically significant association of duration and hemoglobin concentration, platelets, neutrophils counts, there were no interaction observed between addict with other substances and Hb, platelet, neutrophils counts.

Prolonged use also associated with poor educational attainment and high rates of unemployment.

4.3 Recommendations

1- THC and drugs examination test could be used as early diagnosis of anxiety, panic attacks, and psychotic symptoms, in addicted cannabis people.

2-Raising educational awareness, and level.

3-Reduce unemployment rate.

4-Further investigations are recommended with larger sample size and using of more quantitative test.

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Appendix

جامعة السودان للعلوم والتكنولوجيا

كلية الدراسات العليا

استبيان

١. الرقم المتسلسل ..... ٢. العمر .....

٣. المؤهل التعليمي:

( ) أساس ( ) ثانوي ( ) جامعي ( ) مؤهل تقني

٤. طبيعة العمل:

( ) دائم ( ) مؤقت

٥. هل تتعاطى الحشيش؟

( ) نعم ( ) لا

٦. إذا نعم ما هي طريقة التعاطي؟

( ) استنشاق ( ) خلطه مع الطعام ( ) تبخير

٧. ماهي مدة التدخين؟

( ) أقل من سنة ( ) أقل من ٥ سنة ( ) أقل من ١٠ سنة

٨. هل تتعاطى مواد أخرى؟

( ) سجائر ( ) تمباك ( ) شيشة ( ) كحول

٩. هل تعاني من أي مرض؟

( ) مزمن ( ) فيروسي ( ) بكتيري ( ) طفيلي

١٠. إذا نعم ما مدى حدة المرض؟

( ) حاد ( ) وسط ( ) خفيف

Appendix

Table 3.1 The distribution of subject according to addict.

Addict	Frequency	Percent
Yes	40	67
No	20	33
Total	60	100

Table 3.2 The distribution of subject according to their age

Age	Frequency	Percent
18-20	11	18.3
21-23	11	18.3
24-26	10	16.7
27-29	13	21.7
30-32	7	11.7
33-35	7	11.7
36-38	1	1.7
Total	60	100

Table 3.6 The distribution of subject according to their nature of job

Nature of Job	Frequency	Percent
Permanent	17	43
Causal	23	57
Total	60	100

Table3.7 Distribution of cannabis addicts according to duration

Duration	Frequency	Percent
0-2	7	17.5
3 to 5	23	57.5
6 to 8	6	15
9 to 11	4	10
Total	40	100

SYSMEX-KX-21N

CANNABIS