Pharmacodynamics of Secondary Metabolites Extracts of Some Plants from Cholistan Desert in Altering in Vitro Human Haematological Indices

Ghulam Yasin*, Iqra Anwer, Iqra Majeed, Mubashrah Sabir, Sundas Mumtaz, Amna Mehmood

Department of Botany, Bahauddin Zakariya University, Multan, Pakistan.

ABSTRACT

Background: The vegetation of deserts comprise of plants synthesizing secondary metabolites, which have phytopharmacological potential and provide a mechanism of secondary defense against stresses, pathogens, and herbivores. Secondary metabolites, when used by animals and human beings as food or medicine, also defend the individual against diseases. The trafficking of defense strategy of secondary metabolites can be judged by the health status of an individual when treated with plant resourced metabolites as food or medicine. Blood characteristics of humans determine the physiology and health status of the individual. So any change brought in blood by exposure to secondary metabolites can determine the importance of their transfer from plants to humans. Design: The extracts were mixed with human blood in a 1:4 ratio and were analyzed by using the Automated Hematology Analyzer machine for complete blood count tests (CBC). Data were statistically analyzed by using one way ANOVA (Analysis Of Variance). The level of statistical significance was set at ($P < 0.05$). Means were separated by Duncan's multiple range tests. Results: The results showed that leaves, stem, and flower ethanolic extracts significantly increased leukocyte, granulocyte, granulocyte count, HCT, and MCV while significantly decreased eosinophils, platelets, and RBC. On some of the parameters, different parts of the plant showed different effects on the same hematological parameter as leaves and stem significantly reduced the lymphocyte but flower caused an insignificant reduction. Leaves and stem resulted in a significant increase of monocyte and monocyte count but flower significantly and insignificantly reduced the monocyte and monocyte count, respectively. Leaves and flowers of the plant insignificantly decreased the HGB while stem caused a significant increase. A significant increase of the MCH was observed in the case of leaves and stem while flowers showed an insignificant increase. Leaves and flowers significantly decreased the MCHC and stem insignificantly decreased the MCHC.

Key Words: Pharmacodynamics, secondary metabolite, Cholistan, desert, human, hematological.

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INTRODUCTION

Primary metabolites are widely distributed naturally in all living organisms. In plants, such compounds are often present in high concentrations in seeds and vegetative storage organs and are needed for physiological development because of their role in basic cell metabolism [1]. Primary metabolites are for example; sugars, proteins, lipids, and starch, which play vital roles in the growth of plants.

Secondary metabolites are organic compounds; occur in plants in high structural diversity [2]. These compounds are synthesized in plants tissue or organ in a specific way by specific biosynthetic enzymes [3-5], transported through xylem and phloem and are accumulated and stored in high concentrations in the plant organs that are important for survival and reproduction. Secondary metabolites play different functions such as protection

Corresponding author: Ghulam Yasin
Address: Department of Botany, Bahauddin Zakariya University, Multan, Pakistan.
E-mail: ✉ yasingmn_bzu @ yahoo.com
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against high radiation, extreme temperatures, pathogens, and drought conditions. Drought and nutrients scarcity are the main problems faced by the vegetation of the desert area [6]. In deserts, the survival of plants is subjected to the adaptation of particular characters. This limits the species of a plant adapted to the desert environment [7-11]. The Cholistan, in Pakistan, is the world's seventh-largest desert. The climate of the desert is hot and the soil is sandy in texture with negligible amounts of organic matter [12]. Mean summer temperature varies from 35 to 50ºC and winter from 15 to 20ºC, annual rainfall is less than 100 mm to 250 mm [13]. Due to which, the vegetation of the Cholistan desert comprises xerophytic species, which are adapted to these various environmental stresses, like extreme aridity, salinity, high temperature, and low nutrient availability [14]. The topography of soil and its chemical composition determines the vegetation type and plant distribution in that area [15, 16].

Antioxidative nature of secondary metabolites enables them to be used as medicines and food for curing since a long time ago [17, 18]. Even today, plants and their products are fundamental units of health care [19]. Plant extract-based medicine is culturally well accepted by rural and even urban peoples of the world. Medicinal potential, antioxidant nature, and toxicity of the plant resourced secondary metabolites can be assessed for the shift of their defensive role from plant to human. Assessment of human blood parameters reveals the health status of an individual. This is because blood plays a vital role in the nutritional, physiological, and pathological status of the organism [20]. Among these, physiological parameters could be a valuable means of diagnosing a disease [21]. Hence, the investigation of various hematological parameters might be a useful index that can be employed to assess the transfer of secondary immunity from plants to human beings as defensive strategies against stress or disease. Toxic effects of herbal extract in animals are used to deduce potential health risks for humans indirectly [22]. However, direct evaluation of the toxic effects of herbal extracts on human hematological parameters can have more predictive value for human health. The present experiment was designed to explore the trafficking of secondary metabolites from plants to humans as secondary defensive strategies against stressful agents.

MATERIALS AND METHODS

The choice of investigations

The choice of our experiment aimed to explore the relationship between biochemical defense strategies of plants and humans as consumers. Ethanol soluble secondary metabolites [23] sourced from desert vegetation, for in vitro changes in human blood characteristics, can reveal a correlation of biochemical defense of producers and consumers. The assessment of the effects of secondary metabolites on humans discriminating the defensive and toxicological validation of plants' secondary metabolites [24] will provide profound bases to find the trafficking of defense from producer to consumers. The choice of in vitro utilization of human blood is based on the fact that the assessment of blood parameters reveals the health status of an individual. This is because blood plays a vital role in the physiological, nutritional, and pathological status of the organism [20]. Among these, physiological parameters are dependent on blood characteristics, which could be a valuable means of diagnosing a disease [21]. Hence, the investigation of various hematological parameters might be a useful index that can be employed to assess the toxic or defensive potentials of plant extracts containing secondary metabolites [25]. Toxic effects of plants' extract are commonly used in animals to deduce potential health risks for humans indirectly [22]. However, in vitro evaluation of toxicity effects on human hematological parameters directly, can have more predictive value for human health. Moreover, the selection of desert plants is based on the criteria, due to arid environmental conditions, these synthesize secondary metabolites for adaptation to a stressful environment [14].

Experimental design

Plants of the Cholistan desert were selected for secondary metabolites extraction. Ethanol was used as a solvent for extraction procedures [26, 27]. Human blood from a healthy volunteer was utilized for in vitro assessment of hematological indices [28, 29]. The mean of three replicates for each sample was used to reduce the error for comparison with normal blood characteristics.

Climate and geology of the experimental site

The climate of the Cholistan desert of Pakistan is hot and the soil is sandy in texture and contains negligible amounts of organic matters [12]. Based on the topography, parent material, soil, and vegetation, the Cholistan desert is divided into two geomorphic regions; northern lesser Cholistan and southern greater Cholistan. The vegetation of the Cholistan desert comprises species adapted to these various stresses, like extreme aridity, high salinity, high temperature, and low nutrient availability [14].

Plant sample collections

After a preliminary survey of the Cholistan desert, the intact specimen was collected and herborized from the study area. Identification of the herborized material was performed by specialists, by matching them with the labeled herbarium exsiccates lying in the departmental

herbarium (Dr.Mumtaz Bukhari herbarium) of Botany Department Bahauddine Zakarya University, Multan Pakistan and/or the literature [30]. Comprehensive field survey and plant specimen collection, after a preliminary visit, was from Cholistan desert being easily approachable, rich floristically, and with a sizable population of desert herbs. Data and specimens were collected according to an appropriate methodology [31, 32] keeping uniformity among the age of plants, size of plants, and size of dunes. Further processing of collected specimens was carried out in the laboratory of the department. The specimens were first washed with water and later with 2% ethanol to remove dust and other surface contaminants, dried at room temperature and were pulverized to a fine powder by using pestle and mortar.

Crude herbal extract preparation

Following the procedure adopted by Afolayan *et al.,* (2010), the crude ethanolic extract was prepared from finely grounded 1.0 g in 10ml of ethanol by shaking at room temperature for 3h [33]. The ethanol used was of the highest purity. The extract was filtered and the residue was re-processed for extraction. The solvent was evaporated by rotatory evaporator at 40^0 C and the material was stored at -4°C.

Blood sampling and in vitro analysis

After ensuring the confidentiality and anonymity of a blood donor and approval from the local ethical committee, human blood was obtained from a healthy volunteer of 25 years of age having O^+ blood group. The volunteer was selected after a questionnaire of not taking any medications or addictive substances (including tobacco, alcohol, and aspirin or any other antiplatelet drugs) and keeping a balanced diet (meat and vegetables) using no antioxidant supplementation. By adding ethanol, the plant extract was diluted up to 5ml. After consulting literature, the ratio of mixing blood to plant extract was determined by a trial method to find an appropriate dose when no coagulation occurred. Finally, the plant extract was added into 4ml blood (1,4) and was shaken smoothly. The blood sample without the addition of extract was considered as a control for comparison. Complete blood count tests (CBC) by using the Automated Hematology Analyzer machine was performed for hematological indices.

Statistical analysis

Data obtained for blood tests were analyzed by using one way ANOVA (Analysis Of Variance) at a 5% level of statistical significance. Means were compared by Duncan's multiple range test [34].

3. RESULTS

Salsola imbricata stem:

Different sensitivity range was found in the response of blood parameters treated with the ethanolic extract. The application of extract seems more promising in enhancing the eosinophils count (95.28%), HGB (21.24%), HCT (8.05%), MCV (21.36%), MCH (34.28%), MCHC (13.39), RDW count (59.56%), platelets (18.22%). But the extract failed to maintain the trend in granulocyte count, leukocytes, lymphocytes, monocytes, RBC, and MPV parameters. The maximum decrease regarding role of extract was for granulocytes count (17692%), leukocytes (90.05%), lymphocyte (24.55%), monocytes (738.26%), granulocyte (250%), lymphocyte count (146.26%), monocytes count (1230.76%), eosinophils count (95%), RBC (12.06%), and MPV (22.65%).

Salsola imbricata root:

The stimulating behavior of the ethanolic extract varied considerably. The expression of mean values solidify the enhancing role of ethanolic extract for eosinophils (89.13%), HGB (15.95%), platelets (29.75%), MCV (18.96%), MCH (48.79%), MCHC (24.48%) and RDW (17.17%). blood parameters, leukocytes (123.80%), granulocyte count (12615.38%), lymphocyte (9.45%), monocytes (1355.61%), granulocyte (253%), lymphocyte count (146.26%), monocyte count (2669.23), eosinophils count (97.28%), RBC (15.95%), HCT (4.71%), and MPV (3.89%) revealed a significant decrease when treated with the extract.

Salsola imbricata fruit:

The stimulating behavior of the ethanolic extract varied considerably. The significant decrease was in leukocyte (63.41%), granulocyte (20615.38%), lymphocyte (14.12%), Monocyte count (639%), granulocyte (250%), lymphocyte count (90.29%), monocyte count (1023.07%), eosinophils count (98%), RBC (12.06%), MCHC (3.66%) and MPV (19.19%). The observations are excluded from the ongoing trends for eosinophils, HGB, HCT, MCV, MCH, RDW, and platelet parameters. Here, it can be significantly discriminated that the extract has played pivotal role in increasing eosinophils (89.13%), HGB (22.44%), HCT (24.76%), MCV (43.67%), MCH (40.11%), RDW (94.16), and platelets (15.57%).

Leptadenia pyrotechnica root:

An expression of percentage differences supported the manipulation of blood parameters by ethanolic extract application. The greatest promising role of extract, if the term may be used, in this fashion, was for lymphocyte (31.14%), eosinophils count (100%), RBC (24.51%), HGB (36.37%), HCT (56.70%), MCV (18.74%), and RDW count (21.80%). The extract did not reveal any

statistically clear cut increase in MCH (3.53%) and monocyte count (100%). The increasing augmentation could not have a pace for some parameters. The extract showed a decrease in leukocyte (80.46%), granulocyte count (51438.4%), granulocyte (673%), lymphocyte count (25.73%), eosinophils count (93%), MCHC (8.66%), platelets (19.51%), and MPV (10.53%).

***Leptadenia pyrotechnica* stem:**

The ethanolic extract is a potent factor in determining the change in blood parameters. The most marked increase was in monocyte (84.69%), eosinophils count (89.13%), eosinophils (100%), HGB (2.04%), HCT (34%), MCV (57.17%), MCH (27.95%), RDW (66.64%), and platelets (29.49%). The application of the extract seems to significantly decrease leukocytes (39.60%), lymphocyte count (88.87%), granulocyte (673%), lymphocyte (154.85%), monocyte (207.69%), RBC (15.75%), and MPV (32.75%). Although not statistically justified, a considerable decrease in MCHC (20.01%) was observed when the extract was applied.

***Prosopis cineraria* Stem:**

The ethanolic extract is a potent factor in determining the change in blood parameters. The most marked increase was observed in leukocyte (98.93%), granulocyte count (21900%), monocyte count (1047.95%), lymphocyte (108.95%), monocyte (1976.23%), eosinophils (100%), HGB (20.48%), HCT (7.62%), MCV (20.99%), MCH (40.99%), MCHC (13.38%), and RDW (70.80%). Ethanolic extract role deviated from these expectations of promotion in MPV, platelets, granulocyte, eosinophils, lymphocytes, and RBC parameters. The application of extract seems to significantly decrease MPV (30.43%), platelets (9.65%), granulocyte (360%), eosinophils (89.28%), lymphocyte (2.92%) and RBC (12.81%).

***Tamarix aphylla* bark:**

The stimulating behavior of ethanolic extract varied considerably. Here, it can be significantly discriminated that the extract has played a pivatol role in increasing leukocytes (50.97%), granulocyte count (28864%), monocytes count (190%), lymphocyte (71.64%), monocyte (284.61%), eosinophils (100.87%), HGB (14.81%), HCT (18.94%), MCV (37.32%), MCH (38.88%), RDW (86.94%), and platelets (79.12%). The observations are excluded from the ongoing trends for MPV, MCHC, granulocyte, eosinophils, and RBC parameters.

***Tamarix aphylla* stem:**

Ethanolic extract proved its marked influence when applied. Expression of mean values solidify the enhancing role of the ethanolic extract for leukocytes (118.47%), granulocyte count (1284.61%), monocyte count (2586.75%), lymphocyte (105.22%), monocyte (5100%), eosinophils (105.96%), HGB (1.56%), HCT (18.94%), MCV (47.84%), MCH (35.52%), RDW (92.77%) and platelets (9.81%). An exception in this correlation was found MPV, RBC, lymphocyte, eosinophils count, and granulocyte parameters. Blood parameters, MPV (20.63%), MCHC (19.78%), RBC (15.53%), lymphocyte count (0.39%), eosinophils count (89.28%), and granulocyte (20%) revealed a significant decrease when treated with extract.

***Capparis decidua* stem:**

Ethanolic extract substantially altered blood parameters. Exposure to extract has stronger impact of leukocytes (81.70%), granulocyte (2769.23%), lymphocyte count (14.67%), monocyte count (971.42%), lymphocyte (107.46%), Monocyte (1715.38%), Eosinophils (105.96%), HGB (14.76%), HCT (5.58%), MCV (38.45%), MCH (23.97%), RDW (51.42%) and platelets (7.47%) respectively. Ethanolic extract affected MPV, RBC, eosinophils count, and granulocyte with different degrees thereby lowering values from control. Blood parameters MPV (20.63%), RBC (18.87%), eosinophils count (98.92%), and granulocyte (346%) revealed a significant decrease when treated with the extract. Although not statistically justified, but a considerable decrease was observed in MCHC (20.49%) when the extract was applied.

DISCUSSION

The results revealed a diversified action of ethanolic extracts sourced from different plant parts on various hematological attributes. The results are in harmony with the earlier findings of Swenson and Reece, 1993; Olson *et al*, 1984; Ganong, 1997; and Straus, 1998, while in contradict to the findings of Lohar *et al*, (2009) and Gruchy, (1976) regarding RBC and Hb concentration (MCH, MCHC) [21, 35-39]. According to the opinion of some researchers, some herbal plants are often non-specific in their actions [40]. Many plants have been known to produce biologically active substances, some of which are related to special flavor or taste and others are found to be useful as antioxidants. Among the dietary phenolic compounds, antioxidants and secondary metabolites of plants are the most abundant natural antioxidants [41]. Antioxidants act as cell saviors in a number of ways; as reducing agents, free radical scavengers, singlet oxygen quenchers, or hydrogen donors [42]. Antioxidants are synthesized in plants under environmental stresses to neutralize the activity of reactive oxygen species (ROS). Damage to cells and biomolecules caused by reactive oxygen/nitrogen species

(ROS/RNS) is decreased by antioxidants [41]. The plant extracts usually have a high concentration of antioxidants like flavonoids and phenolic compounds. As potent antioxidants, flavonoids are especially important for protection against human diseases. [43]. The concentration ratio of antioxidants to ROS determines not only the potential of plants to withstand adverse environmental conditions but when present in plant extract or its products, these might play important roles against the hemolytic activity of ROS by stabilizing blood cells and molecules or by their direct action on ROS.

Red Blood Cells (Erythrocytes) are considered to be the most abundant cells in the human body, possessing desirable physiological and morphological characteristics. Erythrocytes are affected more than other blood cells by medicines [44].

Results revealed that the ethanolic extracts of leaf, stem, and flower sourced from various plants reduced the RBC (Table 1). Erythrocytes are considered as a major target of free radicals (ROS) owing to the presence of high concentrations of membrane polyunsaturated fatty acids [45]. Oxidative damage to the erythrocyte membrane may be due to the hemolytic activity of ROS. The hemolysis of red blood cells by ROS destructs cell membranes with the release of hemoglobin from these cells. These factors cause deterioration of cell membranes, which may be the key episode of the lysis of cells [46].

Results revealed that the extracts of leaf, stem, and flower of some plants decreased MCHC and increased MCH and MCV (Table 1). MCHC and MCH are indices of hemoglobin concentration in blood and in each cell, respectively [47] and mean cell volume (MCV) refers to the volume of each red cell [48]. RBC and Hb are associated with the total numbers of red blood cells while MCV, MCH, and MCHC relate to individual red blood cells [22]. An increased level of MCV accompanied by a reduction in MCHC depicts the reduction in osmotic fragility of the cell membrane [49]. MCV determines the size of RBCs. RBCs of normal size are termed normocytic [15]. When the MCV is high, RBCs will be larger than normal and are termed as macrocytic. When the MCV is below normal, the RBCs will be smaller than normal and are called as microcytic. These categories of size are used to classify anemias. A significant reduction in MCV, if observed, accounts for interference in iron uptake by hemoglobin. Furthermore, it is reported that a decrease in the blood cells may be due to the increased glycosylation (nonenzymatic) of membrane proteins, which can cause hyperglycemia. Oxidation of glycosylated membrane proteins and hyperglycemia causes an increase in the production of lipid peroxides which, may cause hemolysis of cells.

An observed change in hemoglobin concentration might be due to iron deficiency as iron is an essential component of hemoglobin. Iron deficiency could be due to the direct interference of molecules with iron or interference in its metabolism [19]. ROS has been reported to cause mobilization of Fe^{2+} by Ca^{2+} via Fenton reaction thereby creating iron deficiency [50]. Hemoglobin deficiency results in cells smaller than the normal size. This occurs in many diseases including iron deficiency anemia, thalassemia, and anemias associated with chronic infection or disease.

An increase in hemoglobin (MCH and MCHC) facilitates oxygen transport to the tissues [39]. Increase in hemoglobin concentration may be due to the presence of active principles that stimulate hemopoiesis, or support in the availability of iron for hemopoiesis, or agents for chelating iron may be weakly present or completely absent in the plant extract, which induces a change in hemolysis of RBC [38]. Change in blood platelets might be by their adhesion to collagen and aggregation with ROS species in resting blood platelets or thrombin activated platelets [51]. Variation in the reduction of blood platelet might also be by the antiplatelet activity of antioxidants [52]. Therefore, the compounds that have antioxidative activity might inhibit platelet function. Platelet reduction may be beneficial at some level because platelets reduce the blood viscosity lowering its pressure and may be beneficial in terms of clinical hematology. An increase or decrease in blood attributes might be owed to the anticoagulation activity of the plant extract [28]; free radical scavenging activity of extract [27]; antiglycosylation [53]; by genotoxicity [54] or by thrombolytic potential [29].

CONCLUSION:

Plants revealed dynamics in their phytochemical potential for secondary metabolites. Intense care and extensive work are needed for exploring their ethnomedicinal importance.

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Table 1. In vitro effect of the Ethanolic extract of shrubs of drawer fort on human hamatology [values represent mean±standard deviation; n=3]

Name of species	Total leukocyte (10×3/u L)	Granulocyte (%)	Lymphocyte (%)	Monocyte (%)	Eosinophils (%)	Granulocyte (10×3/L)
Normal blood	5.63 ± 0.05 K	0.13 ± 0.05 o	47.93 ± 0.05 İ	1.96 ± 0.05 m	2.76 ± 0.05 a	0 ± 0 d
***Salsola imbricata(r)* LSD= 2.08**	12.6 ± 0.5 d (-123.80)	16.53 ± 0.92 k (-12615.38)	52.46±1.30 bcde (-9.45)	28.53 ± 1.06 c (-1355.61)	0.3 ± 0.1 1 bc (+89.13)	2.53 ± 1.02 c (-253)
***Salsola imbricata (s)* LSD= 2.97**	10.7 ± 0.55 ef (-90.05)	23.13 ± 1.106 i (-17692)	59.7 ± 1.11 b (-24.55)	16.43 ± 0.90 h (-738.26)	0.13 ± 0.05 de (+95.28)	2.5 ± 0.1 c (--250)
***Salsola imbricata (f)* LSD=5.105**	9.2 ± 0.7 gh (-63.41)	26.93 ± 0.60 h (-20615.38)	54.7 ± 1.11 bcd (-14.12)	14.5 ± 0.75 i (-639)	0.3 ± 0.1 bc (+89.13)	2.5 ± 0.1 c (-250)
***Leptadenia pyrotechnica (r)* LSD= 2.49**	10.16 ± 0.50 efg (-80.46)	67 ± 1.5 a (-51438.4)	33 ± 1 h (+31.14)	0 ± 0 m (+100)	0 ± 0 e (+100)	6.73 ± 0.50 ab (-673)
***Leptadenia pyrotechnica (s)* LSD=3.26**	7.86 ± 0.35 ij (-39.60)	0 ± 0 o (+100)	90.53 ± 1.06 a (-88.87)	5.46 ± 0.40 k (+84.69)	0.3 ± 0.1 bc (+89.13)	6.73 ± 0.50 ab (-673)

Values sharing the different letters represent a significant difference in a respective row; values in parenthesis represent percentage difference over control group; LSD= least standard deviation; r=root; s=stem; f=flower

Table 1: (Cont…) In vitro effect of Ethanolic extract of shrubs of drawer fort on human heamatology [values represent mean±standard deviation; n=3]

Name of species	Lymphocyte count (10×3/L)	Monocyte count (10×3/L)	Eosinophils count (10×3/L)	RBC (10×6/ uL)	HGB (g/dL)	HCT (%)
Normal blood	2.68 ± 0.02 j	0.13 ± 0.05 G	50.33 ± 0.57 A	5.14 ± 0.005 ab	8.33 ±0.05 d	22.73 ± 0.05 h
***Salsola imbricata (r)* LSD=2.09**	6.6 ± 0.2 bcd (-146.26)	3.6 ± 0.5 b (-2669.23)	1.45 ± 0.02 fg (-97.28)	4.32 ± 0.57 bc (-15.95)	9.66 ± 0.97 abc (+15.96)	21.63 ± 1.15 gh (-4.71)
***Salsola imbricata (s)* LSD=2.97**	6.8 ± 0.96 bc (-153.73)	1.73 ± 0.60 cdef (-1230.76)	2.53 ± 0.35 cdef (-95)	4.58 ± 0.58h bc (-10.89)	10.1 ± 1 ab (+21.24)	24.56 ± 0.87 ef (+8.05)
***Salsola imbricata (f)* LSD=5.105**	5.1 ± 0.4 cdefg (-90.29)	1.46 ± 0.37 defg (-1023.07)	1± 0.5 gh (-98)	4.52 ± 0.42 bc (-12.06)	10.2 ± 5.57 ab (+22.44)	28.36 ± 1.12 c (+24.76)
***Leptadenia pyrotechnica (r)* LSD=2.49**	3.36 ± 0.50 hi (-25.37)	0 ± 0 g (-100)	3.5 ± 0.6 bc (-93)	6.40 ± 1.27 bc (+24.51)	11.36 ± 1.05 a (+36.37)	35.62 ± 1.02 a (+56.70)
***Leptadenia pyrotechnica (s)*LSD= 3.26**	6.83 ± 0.35 bc (-154.85)	0.4 ± 0.2 fg (-207.69)	0 ± 0 h (-100)	4.33 ± 0.61 bc (-15.75)	8.5 ± 0.85 bcd (+2.04)	30.46 ± 0.95 b (+34.00)

Values sharing the different letters represent a significant difference in respective column; values in parenthesis represent percentage difference over the control group; LSD= least standard deviation; r=root; s=stem; f=flower

Table 1: (Cont…) In vitro effect of the Ethanolic extract of shrubs of drawer fort on human hematology [values represent mean±standard deviation; n=3]

Name of species	MCV (FL)	MCH (PG)	MCHC (%)	RDW (%)	Platelets (10×3/uL)	MPV (10×3/L)	RDW
Normal blood	44.13 ± 0.05 k	16.13 ± 0.05 h	36.63 ± 0.05 j	13.71 ± 0.02 l	642.33 ± 0.57 i	6.93 ±0.05 a	17.41 ± 0.02 d

***Salsola imbricata* (r) LSD= 2.09**	52.5±1.25 i (+18.96)	24 ± 0.85 a (+48.79)	45.56±0.87 a (+24.48)	22.56 ± 0.90 fghi (+64.67)	833 ± 6135 f (+29.75)	6.66 ± 0.77 ab (-3.89)	20.4 ± 1.34 bc (+17.17)
***Salsola imbricata* (s) LSD= 2.97**	53.56±1.25 hi (+21.36)	21.66±1.205 cdef (+34.28)	41.7 ± 0.95 b (+13.93)	21.86 ± 1.55 fghij (+59.56)	759 ± 6.65 g (+18.22)	5.36 ± 1.28 bcdef (-22.65)	21.4 ± 1.41 abc (+22.91)
***Salsola imbricata* (f) LSD= 5.105**	63.36 ± 0.73 bc (+43.67)	22.6 ± 1.05 abcd (+40.11)	35.26 ± 1.10 efgh (-3.66)	26.6 ± 0.75 abc (+94.16)	742 ± 7 g (+15.57)	5.6 ± 0.85 bcdef (-19.19)	21.96 ± 1.71 abc (+26.13)
***Leptadenia pyrotechnica* (r) LSD= 2.49**	52.4 ± 1.27 i (+18.74)	16.7 ± 0.85 h (+3.53)	33.43± 1.26 hi (-8.66)	16.7 ± 0.76 k (+21.80)	517 ± 4.50 k (-19.51)	6.2 ± 0.2 abcd (-10.53)	18.7 ± 0.7 cd (+7.40)
***Leptadenia pyrotechnica* (s) LSD= 3.26**	69.36±0.97 a (+57.17)	20.6 ± 1.08 e (+27.9571)	29.3 ± 0.95 j (-20.01)	22.83 ± 1.05 fghi (+66.64)	831.33 ± 8.02 f (+29.49)	4.66 ± 0.70 abcd (-32.75)	20.53 ±1.05 abc (+17.92)

Values sharing the different letters represent a significant difference in a respective row; values in parenthesis represent percentage difference over the control group; LSD= least standard deviation; r=root; s=stem; f=flower

Table 2. In vitro effect of the Ethanolic extract of trees of drawer fort on human hematology [values represent mean±standard deviation; n=3]

Name of species	leukocyte count (10×3/u L)	Granulocyte (%)	Lymphocyte (%)	Monocyte (%)	Eosinophils (%)	Granulocyte (10×3/L)
Normal blood	5.63 ± 0.05 k	0.13 ± 0.05 o	47.93 ± 0.05 i	1.96 ± 0.05 m	2.76 ± 0.05 a	0 ± 0 d
Prosopis cineraria (s)	11.2 ± 0.81 e (+98.93)	28.6 ± 1.006 g (+21900)	46.5 ± 1.15 defg (-2.92)	22.5 ± 1.05 e (+1047.95)	0.3 ± 0.1 b (-89.28)	3.6 ± 1.0.5 c (-360)
Tamarix. aphylla (b)	8.5 ± 0.6 hi (+50.97)	38.93 ± 1.05 d (+28864.15)	53.73±1.001 bcde (+12.17)	5.7 ± 1.05 k (+190)	0.03 ± 0.05 e (-98.92)	3.6 ± 0.17 c (-360)
Tamarix. Aphylla (s)	12.3 ± 0.72 d (+118.47)	1.8 ± 0.55 n (+1284.61)	43.4 ± 0.81 efgh (-9.39)	52.66 ± 1.106 a (+2586.75)	0.3 ± 0.1 e (-89.28)	0.2 ± 0.1 d (-20)
Capparis decidua(s)	10.23 ± 0.41 efg (+81.70)	3.73 ± 0.20 m (+2769.23)	54.93 ± 0.40 bcd (+14.67)	21 ± 1 f (+971.42)	0.03 ± 0.05 e (-98.92)	3.46 ± 1.19 c (-346)

Values sharing the different letters represent a significant difference in a respective row; values in parenthesis represent percentage difference over the control group; LSD= least standard deviation; r=root; s=stem; f=flower

Table 2: (Cont…) In vitro effect of the Ethanolic extract of trees of drawer fort on human hematology [values represent mean±standard deviation; n=3]

Name of species	Lymphocyte count (10×3/L)	Monocyte count (10×3/L)	Eosinophils count(10×3/L)	RBC (10×6/ uL)	HGB (g/dL)	HCT (%)
Normal blood	2.68 ± 0.02 j	0.13 ± 0.05 g	50.33 ± 0.57 h	5.14 ± 0.005 c	8.33 ±0.05 d	22.73 ± 0.05 h
Prosopis cineraria(s)	5.6 ± 1.09 cde (+108.95)	2.7 ± 0.85 bcd (+1976.23)	2.56 ± 1.12 ij (+100.874)	4.49 ± 0.81 bc (-12.81)	10 ± 1 abc (+20.48)	24.43 ± 1.13 ef (+7.62)
Tamarix. aphylla (b)	4.6 ± 1.2 efgh (+71.64)	0.5 ± 0.1 fg (+284.61)	2.56 ± 1.12 jk (+100.874)	4.49 ± 0.83 bc (-12.81)	9.53 ± 1.006 abc (+14.81)	27 ± 1 c (+18.94)
Tamarix. Aphylla (s)	5.5 ± 0.85 cdefg (+105.22)	6.76 ± 0.96 a (+5100)	0 ±0 ijk (+105.96)	4.36 ± 1.13 bc (-15.53)	8.46 ± 1.02 bcd (+1.56)	27.8 ± 1.1 c (+22.46)

Capparis decidua(s)	5.56 ± 0.75 cdefg (+107.46)	2.36 ± 0.90 bcde (+1715.38)	0 ± 0 jk (+105.96)	4.17 ± 0.02 bc (-18.87)	9.56 ± 1.001 abc (+14.76)	24 ± 1 ef (+5.58)

Values sharing the different letters represent a significant difference in a respective row; values in parenthesis represent percentage difference over the control group; LSD= least standard deviation; r=root; s=stem; f=flower

Table 2: (Cont…) In vitro effect of Ethanolic extract of trees of drawer fort on human hematology [values represent mean±standard daviation; n=3]

Name of species	MCV (FL)	MCH (PG)	MCHC (%)	RDW (%)	Platelets (10×3/uL)	MPV (10×3/L)	RDW
Normal blood	44.13 ± 0.05 k	16.13 ± 0.05 H	36.63 ± 0.05 j	13.71 ± 0.02 l	642.33 ± 0.57 i	6.93 ±0.05 a	17.41 ± 0.02 d
***Prosopis cineraria* (s)**	53.36 ± 0.91 hi (+20.99)	22.7 ± 1.006 abcd (+40.99)	41.5 ± 1.3 b (+13.38)	23.4 ± 0.79 efgh (+70.80)	580 ± 10.69 j (-9.65)	4.8 ± 1.1 efg (-30.43)	19.73±1.25 bcd (+13.32)
***Tamarix. aphylla* (b)**	60.56 ± 1.20 cde (+37.32)	22.36±1.61 abcde (+38.88)	34.63 ± 1.35 fghi (-5.38)	25.63± 0.80 bcde (+86.94)	1150 ± 5 c (+79.12)	5.53±0.75 bcdef (-20.20)	17.1 ± 5.61 d (-1.78)
***Tamarix. aphylla*(s)**	65.2 ± 1.014 b (+47.84)	21.86 ± 0.86 bcde (+35.52)	29.36 ± 1.20 j (-19.78)	26.43± 0.85 abcd (+92.77)	705 ± 10 h (+9.81)	5.5±0.86 bcdef (-20.63)	21.83 ± 1.10 abc (+25.38)
***Capparis decidua* (s)**	61.06 ± 0.77 cde (+38.45)	19.96 ± 0.80 fg (+23.97)	29.1 ± 0.75 j (-20.49)	20.76 ± 1.101 hij (+51.42)	690 ± 5 h (+7.47)	4.76 ± 0.61 efg (-31.01)	21.2 ± 1.75 abc (+21.76)

Values sharing the different letters represent a significant difference in a respective row; values in parenthesis represent percentage difference over the control group; LSD= least standard deviation; r=root; s=stem; f=flower