Effects of Ethanolic Extract of *Metracaria chamomela* on Some Physiological Parameters in Male Rabbits

Alia Hussein Ali
Department of Biology, College of Science for Women, Baghdad University, Iraq.
E-Mail: aliahusseinali@yahoo.com
Accepted: 25/3/2015

Summary

The aim of this study is to explain the effect of Ethanolic extract 70% of *Metracaria chamomella* on some physiological parameters in male rabbits. Twelve adult male rabbits were used in this study and were divided equally into two groups: First group was control (C) and received normal saline for four weeks, the second group (Treated group) was intubated orally with ethanolic extract of *Metracaria chamomella* in a dose 70 mg/kg B.W. for four weeks. Blood samples were collected by heart puncture from each animal at the end of experiment. Blood sample was divided into a part for hematological study and a part for biochemical analysis. The value of serum urea, and creatinine were reduced in animals that received *Metracaria chamomella* extract at dose of 70 mg/ kg B.W. as compared with the control group. Significant decrease in serum activity of aspartate aminotransferase alanine aminotransferase and alkaline phosphatase activity was observed in *Metracaria* treated animal as compared with the control group. This study explained that there was significant increase in serum total protein, serum albumin and serum globulin in treated animals as compared with the control group. The value of total cholesterol, serum triacylglycerol, serum low density lipoprotein cholesterol and very low density lipoprotein cholesterol concentration were reduced significantly in animal received *Metracaria chamomella* extract while the high density lipoprotein cholesterol was elevated significantly as compared with control group. While the effect of *Metracaria chamomella* extract on blood picture showed no changes in red blood cell count, hemoglobin concentration and hematocrit value but showed significant decrease in platelet count, and significant increase in white blood cell count as compared with control group. Over all this study explained that *Metracaria chamomella* extract had Reno protective and hypolipidimic effect in male rabbit.

Keywords: M*etracaria chamomella*, Urea, Creatinine, Liver enzymes, Lipid profile, Blood picture.

Introduction

Chamomile *Matricaria chamomilla* (MC) is a medicinal plant species from the asteraceae family. Chamomile has calming and smoothing properties. It is used for nervousness, headaches, anxiety and hysteria. It is anti-spasmodic, laxative, analgesic, carminative properties (1 and 2). It is a highly favored and much used medicinal plant in Folk and traditional medicine. It contains a large groups of therapeutically interesting and active compounds. Sesquiterpenes, flavonoids, coumarins and polyacetylenes are considered the most important constituents (3) *M. chamomella* exercised a marked stimulatory action on the secretory function of liver (4). The other pharmacological properties include anti-inflammatory, anti-septic and sedative. *M. chamomella* applied to wounds slow to heal and for skin eruption and infection such as boils and for inflammation of mouth, throat and the eyes. The coumarins are represented in *M. chamomilla* by herniarin, umbelliferone, and other minor ones (5 and 6). *Matricaria chamomilla* contain glucopyranosyloxy-4-methoxycinnamic acid (GMCA), the glucoside precursor of herniarin, were described as native compounds in chamomile (7). Eleven bioactive phenolic compounds (8) such as herniarin and umbelliferone (coumarin), chlorogenic acid and caffeic acid (phenylpropanoids), apigenin, apigenin-7-O-glucoside, luteolin and luteolin-7-O-glucoside (flavones), quercetin and rutin (flavonols), and naringenin (flavanone) are found in chamomile extract. (9). Therefore, the aim of this study was to explain the effect of ethanolic extract of *Metracaria chamomella* in a dose of 70 mg / Kg B.W. on some hematological and biochemical parameters in male rabbits.

Materials and Methods

Chamomile flowers after grinding the dried flowers the plant material was extracted with 70 % ethanol the extract was filtered and evaporated in vacuum rotatory evaporator to yield extract according to (10). The experiment was conducted at the animal house of Biology Department, College of Science for women/ Baghdad. University. Twelve adult Male rabbits weighting 1000-1250 gm. were used in this study. The animals were housed for two weeks for adaptation, they were housed in cages in a room with controlled temperature and humidity. They were kept under good hygienic conditions. Animals were maintained on a natural 12h light and 12h dark cycle, received a balanced diet, water and libitum throughout the experimental period. Rabbits were divided into two groups (n=6) and treated for four weeks as follows: Control group(C) received normal saline orally daily for four weeks and treated group (T) received orally ethanolic extract of Metracaria chamomella (MC) at a dose of 70 mg/Kg B.W (11). At the end of the experimental period, overnight fasting, blood samples were collected by heart puncture in tubes containing EDTA anticoagulant for hematological study. Serum was separated from coagulant blood by centrifugation at 5000 rpm for 10 minutes and stored at - 20C^{o} for studying the following: Serum urea, creatinine concentration.

According to Diamond enzyme kit (12 and 13), serum aspartate aminotransferase (AST), serum alanine aminotransferase (ALT) and alkaline phosphatase activity (AP) using enzymatic kit (14 and 15). Total proteins were estimated by using Biuret method as described (16), serum albumin gm/dl was estimated by albumin kit (17) and serum globulin concentration g/dL was estimated in directly by measuring of albumin in serum and then it was subtracted from total protein. Determination of serum total cholesterol (TC) concentration using enzymatic assay kit (18), triacylglycerol TAG by using enzymatic assay kit (19) high density lipoprotein cholesterol (HDL-C) concentration using enzymatic assay kit (20), low density lipoprotein cholesterol (LDL-C) concentration and very low density lipoprotein cholesterol (VLDL-C) concentration according to (21). Blood with EDTA anticoagulant used for hematological study. Estimation of hemoglobin (Hb) concentration was according to (22), white blood cell count was according to (23), red blood cells and hematocrit (PCV) using the technique of (24). Total platelets count was according to (25). The data were analyzed using the statistical package for social since program (S.P.S.S.). For comparison between different experimental groups analysis of variance ANOVA was used. The results were expressed as means ± SE and ($P<0.05$) was conceded to be statistically significant (26).

Results and Discussion

The results (Table, 1) showed that oral administration of ethanolic extract of MC for four weeks on serum urea caused significant ($P<0.05$) decrease in the serum urea concentration as compared with control group. As well as, significant ($P<0.05$) decrease in serum creatinine concentration was observed in T1 group as compared with control. A significant ($P<0.05$) decrease in serum AST, ALT and AP activity was recorded in MC treated animals as compared with control group. (Table, 2) There was a significant ($P<0.05$) increase in total serum protein in treated animals as compared with control group with significant ($P<0.05$) elevation in serum albumin and globulin concentration in treated animals as compared with control group. The results in (Table, 3) oral administration of Metracaria chamomella (Mc) alcoholic extract (70 mg / kg B.W.) caused significant ($P<0.05$) decline in serum total cholesterol (T.C.), STAG, LDL-C and VLDL-C concentrations as compared with control group. Besides There was significant ($P<0.05$) increase in serum high density lipoprotein cholesterol concentration (HDL-C) in treated animals as compared with control.

Table (4) oral administration of ethanoic extract Metracaria chamomella 70 mg/ kg for four weeks on blood picture. There were no changes on red blood cell count, hemoglobin concentration, hematocrit value with significant ($P<0.05$) decrease in platelet count as compared with the control group. Also there was a significant ($P<0.05$) increase in white blood cell count in treated group as compared with control.

Table, 1: Effect of ethanolic extract from *Metracaria chamomella* on some serum biochemical parameters in male rabbits.

Parameters / Groups	Serum urea	Serum creatinine	Aspartate amino transaminase (AST)	Alanine amino transaminase (ALT)	Alkaline phosphates
Control (C)	89 ± 0.75 A	1.3 ± 0.30 A	35 ± 1.48 A	41 ± 1.02 A	63 ± 0.79 A
Treated (T)	72.8±1.96 B	0.47±0.44 B	21 ± 2.26 B	19.8 ± 1.98 B	43 ± 1.32 B

Values are expressed as mean ± SE, n=6
T: Rabbits received 70 mg / Kg B.W. of ethanolic extracted from *Metracaria chamomella*
Capital letters denoted difference between groups P<0.05 vs control.

Table, 2: Effect of ethanolic extract from *Metracaria chamomella* (Mc) on serum total protein, albumin and globulin concentrations (gm/dL) in male rabbits.

Parameters / Group	Total protein	Albumin	Globulin
Control (C)	42.40 ± 0.54 B	24.80 ± 0.83 B	17.8 ± 0.62 B
Treated (T)	68.60 ± 0.98 A	32.25 ± 1.14 A	31.6 ± 1.50 A

Values are expressed as mean ± SE, n=6
T: Rabbits received 70 mg / Kg B.W. of ethanolic extracted from *Metracaria chamomella*
Capital letters denoted difference between groups P<0.05 vs control.

Table, 3: Effect of ethanolic extract from *Metracaria chamomella* on Serum lipid profile concentration (mg/dl) in male rabbits.

Parameter / Group	TC	STAG	HDL-C	LDL-C	VLDL-C
Control (C)	71.6 ± 1.22 A	88 ±7.07 A	27 ± 0.38 B	27 ± 1.22 A	17.6 ± 0.70 A
Treated (T)	68.3±2.55 B	66.3±6.17 B	32±0.69 B	22.4 ± 1.89 B	13.6±1.67 B

Values are expressed as mean ± SE, n=6
T: Rabbits received 70 mg / Kg B.W. of ethanolic extracted from *Metracaria chamomella*
Capital letters denoted difference between groups P<0.05 vs control.

Table, 4: Effect of ethanolic extract from *Metracaria chamomella* on some hematological parameters in male rabbits.

Parameters / Group	Red blood cell count X 10^6	Hemoglobin (gm./dl)	PCV (%)	Platelet count X 10^3	White blood cell count X 10^3
Control (C)	5.7±0.11 A	13.2±0.47 A	37.15±3.39 A	1208.75±3.4 A	6.6±1.13 B
Treated (T)	5.8±0.66 A	12.75±1.24 A	36.0±3.16 A	971.5±2.43 B	7.98±1.24 A

Values are expressed as mean ± SE, n=6
T: Rabbits received 70 mg / Kg B.W. of ethanolic extracted from *Metracaria chamomella*
Capital letters denoted difference between groups P<0.05 vs control.

The use of natural products due to natural available is a general trend now (27). The decrease in serum level of urea and creatinine in animals treated with chamomile might be due to the anti-oxidant activity of chamomile (28). In the present study the significant (P<0.05) decrease of serum AST, ALT and AP activity, the assay of these enzymes were important in the diagnosis of liver function and considered as a markers of liver dysfunction (29). The decrease of liver enzymes in animals that received chamomile indicates the hepatoprotective effect of chamomile (4). Liver is an important organ for protein synthesis, so the increase in serum level of total protein, albumin and globulin indicate the hepatoprtective effect of *Metracaria chamomella* (30). *Metracaria chamomella*

contain polyphenolic compounds had benefit by several mechanisms including direct free radical quenching protection and regeneration of liver cells (31) flavonoid accelerate regeneration process and production of liver cells which were responsible for protein synthesis (32). In the current study there were significant decrease in serum TC, TAG, LDL-C and VLD-L with significant increase in HDL-C concentration follows MC treatment indicating its hypolipdemic effect. (33) mentions that chamomella extract contain high content of flavonoid (63.3 %) most of them are apigenin and total phenolic compounds (23.2 %), bio active compounds reported to act as free radical scavenger (34). *Metracaria chamomella extract* as a natural material, lower blood cholesterol by two mechanisms by the presence of flavonoids that enhance the phosphorylation of HMG CoA reductase enzyme indirectly thus diminish endogenous cholesterol production. Also *Metracaria chamomella extract* appeared to protect LDL against oxidation and protected α tocopherol and other endogenous antioxidant in LDL from oxidation (35), which may be useful in alleviating the adverse effects associated with low density lipoprotein LDL cholesterol oxidation in atherosclerosis (36). Besides, (MC) ethanolic extract caused significant elevation in serum HDL-C concentration that play an important role in plasma lipid transport of cholesterol from peripheral cells to the liver for excretiosn and catabolism (37) indicating its anti-atherosclerotic effect .

In this study *Metracaria chamomella* extract contained a flavonoid that declined platelet count which may be due to the inhibition of arachidonic acid metabolism by cyclooxygenase (38). Flavonoids of *Metracaria chamomella* extract (apigenin) were potent inhibitor for platelet aggregation, also *Metracaria chamomella* ethnolic extract had anti-oxidant effect (39), particularly (MC) had anti-thrombotic effect. It was direct scavenging free radical by maintaining proper concentration of endothelial prostacyclin and nitric oxide (40).

References

1. Crotteau, C. A.; Wright, S.T. and Eglash, A. (2006). Clinical inquiries. What is the best treatment for infants with colic? J. Fam. Pract., 55: 634-636.
2. Ganzera, M.; Schneider, P. and Stuppner, H. (2006). Inhibitory effects of the essential oil of chamomile (Matricaria recutita L.) and its major constituents on human cytochrome P-450 enzymes. Life Sci., 78: 856-861.
3. Pirzad, A.; Alyari, M. R.; Shaliba, S.; Zehtab-Salmasi and Moammadi, A. (2006). Essential oil content and composition of German chamomile (*Matricaria chamomilla* L.) at different irrigation regimes. J. Agron., 5: 451–455.
4. Franz, C. H.; Bauer, R.; Carle, R.; Tedesco, D.; Tubaro, A. and Zitterl-Eglseer, K. (2005). Study on the assessments of plants/herbs, plant/herb extracts and their naturally or synthetically produced components as additives for use in animal production. 1: 155–169.
5. Bottcher, H. and Gunther, I. (2005). Chamomile industrial profiles. Boca Raton: CRC Press. Storage of dry drug. Pp: 211–213.
6. Kotov, A. G.; Khvorost, P. P. and Komissarenko, N. F. (1991). Coumarins of *Matricaria recutita.* Khim Prirod Sojed., 6: 853–854.
7. Ohe, C.; Miami, M.; Hasegawa, C.; Ashida, K.; Sugino, M. and Kanamori, H. (1995). Seasonal variation in the production of the head and accumulation of glycosides in the head of *Matricaria chamomilla*. Acta Horticulture., 390: 75–82.
8. Gupta, V.; Mittal, P.; Bansal, P.; Khokra, S .L. and Kaushik, D. (2010). Pharmacological potential of *Matricaria recutita*-A review. Int. J. Pharm. Sci. Drug Res., 2: 12–16.
9. Pino, J. A.; Bayat, F.; Marbot, R. and Aguero, J. (2002). Essential oil of *Chamomilla recutita* (L.) Rausch. From Iran. J, Essent. Oil Res., 14: 407–408.
10. Harborn, J. B. (1984). Methods of extraction and isolation, phytochemical methods, 2nd edition, London, New York champan and hall. Pp: 4-16.
11. Zafari, Z. F.; minaee, B.; Amir zaragar, A. and Mousavi zadeh, K. (2010). Effect of

chamomile extract on biochemical and clinical parameters in arat model of poly cystic ovary syndrome. J. Reprod. Infert. 3:169-174

12. Palton, C. J. and Crouch, S. R. (1977). Enzymatic determination of serum urea by modified Berthelot reaction. Anal Chem., 49: 464-469.
13. Henry, R. J. (1974). Principles and Techniques. Clinical Chemistry, 2nd Ed. Harper and Row. P: 525.
14. Reitman, S. and Frankel, S. (1975). A colourimetric method for the determination of serum glutamic oxaloacetic and glutamic pyruvic transaminases. Am. J. Clin. Path., 28: 56.
15. Belfield, A. and Goldberg, D. M. (1971). Revised assay for serum phenyl phosphatase activity using 4-amino-antipyrine enzyme., 12: 561-573
16. Doumas, B.T. (1975). Standards for total serum protein assays. A collaborative study. Clin. Chem., 21(8): 1159 – 1161.
17. Young, D. S. (1995). Effect of drugs on clinical laboratory tests. 4th ed. AACC. Press
18. Ellefson, R. and Garaway, W. (1976). Lipid and lipoproteins. In: Fundamental of clinical chemistry. Tietz, N.W. (Ed) chap 0. Pp: 512-514. Saunders. W. B. company. Philadelphia, London, Toronto.
19. Rojkin, M. L.; Olguin, D. E.; Mariani, M. C.; Drappo, G. A. y. and Sosa, C. P. (1974). Proteinastotales del sureo: causes mas Frecuentes de error en la roaccion del Biuret Nuevo reactivo cupraol calino estable. Biog. Del. Atlantico, Pp: 1163 – 1193.
20. Bucolo, G. and David, H. (1973). Quantitative determination of serum triglycerides by use of enzymes Clin. Chem., 19: 476-482
21. Friede wald, W.; Levy, X. and Fredrickson (1972). Estimation of the concentration of low density lipoprotein cholesterol in plasma without use of preparation ultra-centrifugation. Clin. Chem., 18: 499–502.
22. Van kampen, E. J. and Zulstra, W.G. (1961). Standerdization of hemoglobinometr: The hemoglobin cyanide method. Clin. Chem. Acta., Pp: 538 – 540.
23. Harris-young, L. (1995). Principles of hematology. Brown publishers. WMX UK. Pp: 145-154.
24. Rodak, F. P. (1995). Routin laboratory evaluation of blood cells and bone marrrow. In: Diagnostic hematology, W. B. sounders Comp. Phild. London, Toronto, Montreal, Sydney, Tokyo. Pp: 125 – 129.
25. Voigt, G. L. (2000). Hematology techniques concepts for veterinary technician 1st ed. lowa State University Press. Pp: 28-52.
26. Daniel, W. W. (1974). Biostatics: A Foundation for analysis in the health sciences. John Wiley and Sons, New York USA. Pp: 157-168, 247-262.
27. Qattan, K. A.; Thomson, M. and Muslim, A. (2008). Garlic (Allium sativum) and ginger (Zingiber officinale) attenuate structural nephropathy progression in Streptozotocin-induced diabetic rats. e-SPEN, the European e-J. Clin. Nutr. Metabol., 3: 62-71.
28. Haidara, M. A.; Mikhailidis, D. P.; Rateb, M. A.; Ahmed, Z. A.; Yassin, H. Z. and Ibrahim, I .M. (2009). Evaluation of the effect of oxidative stress and vitamin E supplementation on renal function in rat with Streptozotocin-induced type 1 diabetes. J. diabetes and its complications. 23: 130-136.
29. Jasmine, R. and Daisy, P. (2007). Hypoglycemic and Hepatoprotective Activity of Eugenia jambolana in Streptozotocin-Diabetic Rats. Inter. J. Biol. Chem., 1(2): 117-121.
30. Andereadou, L.; Efstations, K.; Liodromitis, K.; Maria, C. and Agalias, A. (2006). Constituent oleurppein exhibits ant ischemic, anti-oxidative and hypolipidmic effect in anasthetized rabbits. J. Nutr., 136(14): 2213-2219.
31. Genkinger, J. M.; Platz, E. A.; Hoffman, S. C.; Comstock, G. W. and Helzlsouer, K. J. (2004). Fruit, vegetable, and antioxidant intake and all –case, cancer, and cardiovascular disease mortality in a community – dwelling population in

Washington county, MD. Am. J. Epidemiol., 160:1223-1233.

32. Amic, D.; Beslo, D. and Trinajstic, N. (2003). Structure radical scavenging activity relationships of flavonoids croat chem. Act., 76:55-61.

33. Gupta, A. and misra, N. (2006). Hepatoprotective activity of aqueous ethanolic extract of chamomile capitule in paracetamol intoxicated albino rats. Am. J. Pharm. Toxico., 1: 17-20.

34. Avallone, R.; Zenoli, P.; Pula, G.; Kleinschnitz, M.; Schreier, P. and Barastdi, M. (2000). Pharmacological profile of apigenin, aflavonoid isolated from *Metracaria chamomella* Brochem. Pharmcol., 13(11):1387-1394.

35. Aria, Y.; Watanabe, S. and kimira, M. (2000). Dietry intake of flavonols, flavones and isoflavones by japanes women and the inverse correlation between quercetin intake and plasma LDL cholesterol concentration J. Nut., 130: 2243-2250.

36. Stocker, R. M. and keaney, J. F. (2004). Role of oxidative modification in atherosclerosis, physiological review. 84: 1381-1478.

37. Miller, N. E. (2011). HDL metabolism and its role in lipid transport, Europe Heart J., 32: 20.

38. Tapas, A. R.; Sakarkarl, D .M. and Kakde, R. B. (2008). Flavonoids as nutraceuticals: Tropical J. Pharm. Res., 7(3):1089-1099.

39. Namvaran –abbas-abed, A. and Khayat-nouri, M. (2011). Interactions between matricaria recutita and cisplatin on PTZ-induced seizure threshold in mice. Feyz J. Kashan Uni. Med. Sci., Pp: 15.

40. Gryglewski, R. J.; Korbut, R. and Robak, J. (1987). On the mechanism of anti-thrombotic of flavonoids Biochem. Pharm., 36: 317-322.

تأثير المستخلص الكحولي الأثيلي لنبات البابونج *Metracaria chamomella* في بعض المعايير الفسلجية لذكور الارانب

عالية حسين علي

قسم علوم الحياة، كلية العلوم للبنات، جامعة بغداد، العراق.

E-Mail: aliahusseinali@yahoo.com

الخلاصة

هدفت الدراسة لمعرفة تأثير المستخلص الكحولي 70% لنبات البابونج *Metracaria chamomella* وبجرعة 70ملغم/ كغم من وزن الجسم في بعض المعايير الفسلجية لذكور الأرانب. استعملت في التجربة اثناعشر من ذكور الأرانب البالغة والتي قسمت إلى مجموعتين مجموعة السيطرة والتي أعطيت المحلول الملحي الفسلجي فمويا ولمدة أربعة اسابيع والمجموعة الثانية وهي مجموعة المعالجة والتي أعطيت المستخلص الكحولي لنبات البابونج وبجرعة 70ملغم/ كغم من وزن الجسم لمدة أربعة اسابيع وفي نهاية التجربة سحبت عينات الدم من القلب لمجموعتي السيطرة والمعالجة. جزء من الدم وضع في انابيب حاوية على مانع تخثر لدراسة مؤشرات الدم والجزء الأخر فصل منه مصل الدم للدراسة البايوكيميائية. وقد أوضحت نتائج الدراسة حصول انخفاض معنوي في مستوى اليوريا والكرياتنين والأسبرتيت امينوترانسفيريز والألنين أمينوترانسفيريز والفوسفتيز القاعدي بالمقارنة مع مجموعة السيطرة كذلك حصول ارتفاع معنوي بمستوى بروتينات بلازما الدم والتي شملت مستوى البروتينات الكلية وهي الألبومين والكلوبيولين بالمقارنة مع مجموعة السيطرة. كما بينت النتائج حصول انخفاض معنوي في مستويات الدهون في الدم والتي شملت الكوليسترول الكلي وتراي أسيل كليسرول بلازما الدم ولايبوبروتين-كوليسترول بلازما الدم منخفض الكثافة ولايبوبروتين-كوليسترول بلازما الدم منخفض الكثافة جداً مع حصول ارتفاع معنوي في مستوى اللايبوبروتين-كوليسترول عالي الكثافة في المجموعة المعاملة بالمستخلص الكحولي لنبات البابونج بالمقارنة مع مجموعة السيطرة. أما تأثير نبات البابونج في مؤشرات الدم فلم تحدث زيادة معنوية لعدد كريات الدم الحمراء وحجم خلايا الدم المضغوطة او المرصوصة ونسبة الهيموكلوبين في الدم مع حصول انخفاض معنوي في عدد الصفيحات الدموية وارتفاع معنوي في عدد خلايا الدم البيض بالمقارنة مع مجموعة السيطرة. فضلاً عن أن المستخلص الكحولي لنبات البابونج له تأثير خافظ للدهون ومنشط للكلى في ذكور الأرانب.

الكلمات المفتاحية: نبات البابونج، اليوريا، الكرياتنين، انزيمات الكبد، صورة الدهن، صورة الدم.