Original
ISSN 2413-0516

Mentha piperita and *Thymus vulgaris* Ameliorate Hematological Changes Induced by Chronic Consumption of Oxidized Palm Oil in Rats

Alsufiani Hadeil Muhanna[1,2*], Alsharari Mashael Arif[1,3], Mansouri Rasha A[1], Osman Nadia Nour[1,4]

[1]Biochemistry Department, Faculty of Science, King Abdulaziz University, Jeddah, Saudi Arabia.
[2]Experimental Biochemistry Unit, King Fahad Medical Research Center, King Abdulaziz University, Jeddah, Saudi Arabia.
[3]Chemistry Department, Faculty of Science, Jouf University, Sakaka, Saudi Arabia.
[4]Food Irradiation Research Department, National Centre for Radiation Research and Technology (NCRRT), Cairo, Egypt.
*Correspondence to: Hadeil Muhanna Alsufiani (E-mail:halsufiani@kau.edu.sa)
(Submitted: 01 February 2024 – Revised version received: 20 February 2024 – Accepted: 15 March 2024 – Published online: 26 April 2024)

Abstract

Objective: The aim of this study was to investigate the effect of *Mentha piperita* L and/or *Thymus vulgaris* in ameliorating the hematological changes induced by chronic consumption of oxidized palm oil (OPO) in rats.

Methods: Fifty rats were divided into five groups as follows: rats received a basal diet (control), rats received a basal diet sustained with OPO, rats fed OPO and received *Mentha piperita* extract, rats fed OPO and received *Thymus vulgaris* extract, and finally rats fed OPO and received both extracts. After 6 weeks, blood samples were withdrawn and a complete blood count test was done.

Results: Rats fed a diet containing OPO showed a significant decrease in red blood cells (RBCs), hematocrit (Hct), mean corpuscular volume (MCV), hemoglobin (Hb), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), lymphocytes (LYM), and platelets (PLTs) accompanied by a significant increase in white blood cells (WBCs), red cell distribution width (RDW), and mean platelet volume (MPV) compared to rats fed a normal diet. In contrast, rats fed on diet containing OPO and the aqueous extract of either *Mentha piperita* or *Thymus vulgaris* showed a significant increase in RBCs, Hct, MCV, Hb, MCH, MCHC, LYM, and PLTs accompanied by a significant decrease in RDW, WBCs, and MPV relative to rats fed on diet containing OPO only. Treatment with a combination of both plants showed better improvements.

Conclusion: Both *Mentha piperita* and *Thymus vulgaris* ameliorate the hematological changes induced by OPO.

Keywords: Oxidized palm oil, *Mentha piperita*, *Thymus vulgaris*, hematological indices, antioxidant

Introduction

Cooking oils are an integral part of human diets as they are used in almost all types of culinary practices, including baking, sautéing, dressing, and frying.[1] Frying is a process of immersing food in hot oil that is held at a temperature of 150–190°C, where simultaneous heat and mass transfer occur between the oil and fried food.[2] Palm oil is one of the most-used cooking oils for deep frying purposes. Palm oil is thermally oxidized when the fresh form is subjected to several rounds of heating at high temperatures.[3] Therefore, it is susceptible to oxidative changes because its unsaturated fatty acid constituents readily undergo oxidation resulting in the formation of peroxides, aldehydes, ketones, aldehydroesters, and ozonides.[4] In addition, it is susceptible to the loss of carotenoid, phenolics, and vitamins, thus reducing the overall antioxidant properties of the oil.[1-5]

Several studies have revealed that chronic consumption of oxidized palm oil (OPO) may predispose consumers to various disease conditions thereby having deleterious effects on health. For example, OPO consumption caused growth retardation, thrombosis, fatty liver, adverse lipid profile, essential fatty acid deficiency, a deficiency of nucleic acid, and micronutrient malnutrition.[4] Additionally, OPO induces reproductive toxicity and organ toxicity, particularly of the heart, lung, liver, and kidney.[6] A study by Ani et al. (2015) demonstrated that thermal OPO consumption may be detrimental to the body's hematological system, as it alters packed cell volume (PCV) and white blood cell (WBC), neutrophil, and lymphocyte (LYM) counts.[4] Furthermore, Mesembe et al. (2004) observed that chronic consumption of an OPO diet leads to a significant decrease in PCV, hemoglobin (Hb), and red blood cell (RBC) count and a significant increase in WBC count. They concluded that chronic consumption of an OPO diet may result in anemia and leukocytosis in rats.[7]

Medicinal plants have been reported to have potential in protecting and improving the hematological system in different health situations. One of the oldest medicinal plants is the well-known spice and herb *Mentha piperita* (peppermint) from the Lamiaceae family.[8] *Mentha piperita* oil was found to protect the hematopoietic system against lethal gamma radiation in rats.[9] Moreover, treatment of rats with peppermint essential oil rectified anemia resulting from diabetes and increased counts of leukocytes and platelets (PLTs).[10] These results were further corroborated by Zangeneh et al., (2018) who found that treatment with aqueous extract of *Mentha piperita* leaves significantly enhanced the decrease in RBCs, Hb, mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), and PLTs in rats with ethanol-induced gastroduodenal ulcers as compared to the untreated group.[11]

In addition to *Mentha piperita*, *Thymus vulgaris* (thymus) is the most famous medicinal plant from the Lamiaceae family and has attracted the attention of many investigators for some time. The administration of *Thymus vulgaris* in rats increased the reduced RBCs, Hb, MCHC, and LYMs caused by consumption of thermally oxidized sunflower oil.[12] Moreover, Shittu et al. (2013) found that thymus extract improved RBC count and Hb concentration in Trypanosoma brucei–infected rats when compared to the untreated group.[13] Similar findings were reported by Nada et al. (2013), who found that thymus oil was efficient in ameliorating the alteration in RBCs, hematocrit (Hct), Hb, and WBCs observed in rats exposed to γ-radiation.[14] Thus, numerous investigations have been conducted

on the effects of *Mentha piperita* or *Thymus vulgaris* on the hematological system in different health situations. However, little attempt has been made to investigate the effect of these plants on the hematological system in rats fed OPO. Thus, the aim of this study was to investigate the effect of *Mentha piperita* L and/or *Thymus vulgaris* in ameliorating the hematological changes induced in rats by chronic consumption of OPO.

Materials and Methods

Raw Materials

Palm oil, leaves of *Mentha piperita* L, and *Thymus vulgaris* were purchased from a local traditional market in Jeddah, Saudi Arabia.

Preparation of Oxidized Palm Oil (OPO)

OPO was prepared by frying 1 kg of potatoes in 2.5 L of palm oil for 10 minutes at 180°C ten times. The cooling time between intervals of the heating process was 5 hours. In each frying process, a fresh batch of potatoes was used without adding fresh oil to compensate for oil loss. The test diets were then formulated by mixing 15% (w/w) OPO with commercial rat feed.[15]

Preparation of Mentha Piperita Aqueous Extract

Ten grams of dried leaves of *Mentha piperita* were dissolved in 100 mL of distilled water for a day. Then, the water extract was filtered into an amber bottle and kept at −80°C until further use. The extract was thawed and allowed to stand at room temperature for about 2 hours before use.[16]

Preparation of Thymus Vulgaris Aqueous Extract

Thirty grams of dried leaves of *Thymus vulgaris* were dissolved in distilled water (60 mL) for 1 day. Then, the sample was filtered using filter paper, and this filtrate was stored at 20°C for 3 days only (i.e., freshly prepared every 3 days).[17]

Experimental Animals

Fifty 2 months old adult male Wistar rats weighing 150–200 g were obtained from the Central Animal House, King Fahad Medical Research Center, King Abdulaziz University, Jeddah, Saudi Arabia. The rats were kept in plastic cages and maintained at 25 ± 2°C with a 12 h light-dark cycle. The rats were kept under observation for 2 weeks before the start of the experiment. Animal procedures were performed with the approval (number 581-17) of the Ethics Committee of the King Fahad Medical Research Center and according to recommendations for the proper care and use of laboratory animals.

Experimental Design

Fifty animals were used and divided into five groups (ten rats in each) as follows: Negative control group (Control): Rats received a basal diet; Positive control group (OPO): Rats received a basal diet sustained with 15% (w/w) OPO for 6 weeks;[18] OPO and mentha group (OPO+MEN): Rats fed OPO and received water extract of mentha (290 mg/kg BW/day)[19] for 6 weeks; OPO and thymus group (OPO+THY): Rats fed OPO and received water extract of thymus (500 mg/kg BW/day)[20] for 6 weeks; finally, OPO and mixture of mentha and thymus group (OPO+MIX): Rats fed OPO and received both water extracts (mentha 290 mg/kg and thymus 500 mg/kg) by gavage, respectively, for 6 weeks.

Determination of Hematological Parameters

At the end of the experiment (6 weeks), the rats fasted overnight for scarification. Blood samples were withdrawn by a heparinized capillary tube from the retro-orbital plexus of each rat under anesthesia with diethyl ether. RBCs, Hct, MCV, red cell distribution width (RDW), Hb, MCH, MCHC, WBCs, LYMs, PLTs, and mean platelet volume (MPV) were measured on a Cell-Dyn Emerald 22 Hematologic Analyser (Abbott, USA).

Statistical Analysis

One-way analysis of variance (ANOVA) followed by LSD multiple comparison test were used to assess differences in the hematological parameters between the five groups, and the results are expressed as mean ± standard deviation (SD). All statistical analyses were performed using GraphPad Prism software (version 9.3.1, USA, Biomatters, Ltd. NZ and GSL Biotech, USA). A *P*-value of <0.05 was considered statistically significant.

Results

The effects of the consumption of OPO for 6 weeks on hematological parameters in rats are shown in Figures 1–3. The results showed an alteration in RBC indices characterized by a significant decline in RBC count, proportion (Hct), and average size (MCV) compared to rats fed a normal diet (control group) (Figure 1A, 1B, and 1C, respectively). However, the variability of RBC size (RDW) was higher compared to the control group (Figure 1D). Moreover, Hb concentration, average amount (MCH), and concentration (MCHC) per single RBC were all decreased in rats fed a diet with OPO compared to the control group (Figure 1E–1G, respectively). Results of WBC indices showed an increase in total WBC count, while LYM count was decreased (Figure 2A and 2B, respectively). In contrast to WBC, the PLT count was decreased, whereas their volume (MPV) was increased compared to the control group (Figure 3A and 3B, respectively).

In contrast, the results showed a significant amelioration in hematological parameters among rats fed OPO treated with either mentha (OPO+MEN) or thymus (OPO+THY) compared to untreated rats fed OPO only (Figures 1–3). These treatments resulted in an improvement in the RBC indices characterized by a significant increase in RBCs, Hct, MCV, Hb, MCH, and MCHC compared to untreated rats fed OPO only (Figure 1A–1C, 1E–1G, respectively) and a lower variability in RBC size (RDW) (Figure 1D). In addition to RBCs, WBC and PLT indices were also improved by these treatments. WBC count was decreased, while LYM count was increased (Figure 2A and 2B). Moreover, PLT count was increased, while their volume (MPV) was decreased in comparison with untreated rats fed OPO only (Figure 3A and 3B).

The treatment of rats with the combination of mentha and thymus (OPO+MIX) showed better improvement in the RBC, WBC, and PLT indices when compared with rats fed OPO treated with either mentha (OPO+MEN) or thymus (OPO+THY). Furthermore, the treatment with the mixture showed RBC, WBC, and PLT indices similar to those of rats fed a normal diet (control group) (Figures 1–3).

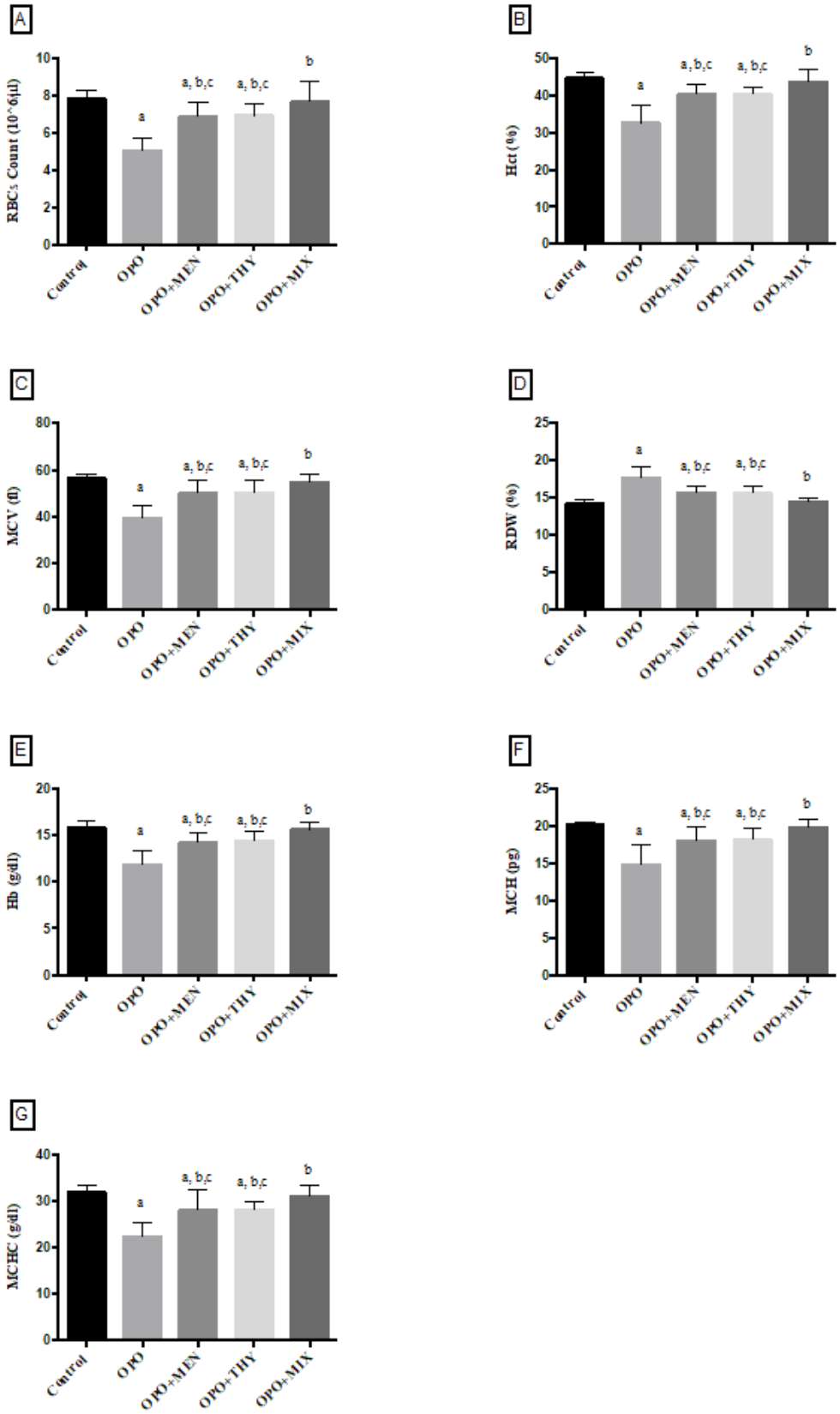

Fig. 1 **Effect of mentha and/or thymus extract on RBC indices [including (A) RBC count (B) Hct (C) MCV (D) RDW (E) Hb (F) MCH (G) MCHC] of rats fed a diet containing 15% OPO for 6 weeks. Data are presented as mean ± standard deviation (SD). Lower-case "a" denotes a significant difference with the control group ($P < 0.05$). Lower-case "b" denotes a significant difference with the OPO group ($P < 0.05$). Lower-case "c" denotes a significant difference with the OPO+MIX group ($P < 0.05$). RBC: red blood cell, Hct: hematocrit, MCV: mean corpuscular volume, RDW: red cell distribution width, Hb: blood hemoglobin, MCH: mean corpuscular hemoglobin, MCHC: mean corpuscular hemoglobin concentration, OPO: oxidized palm oil, MEN: mentha, THY: thymus, MIX; mixture of mentha and thymus.**

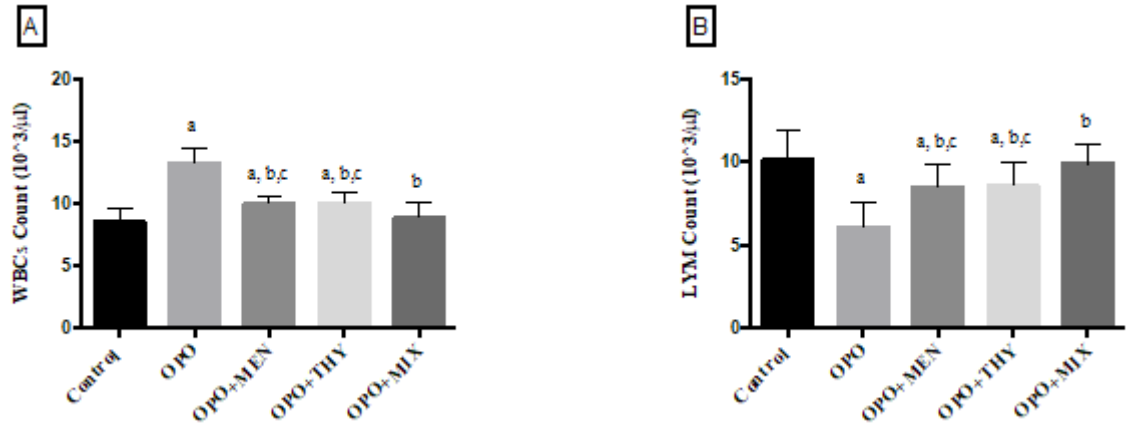

Fig. 2 **Effect of mentha and/or thymus extract on WBC indices [including (A) WBC count and (B) LYM count] of rats fed a diet containing 15% OPO for 6 weeks. Data are presented as mean ± standard deviation (SD). Lower-case "a" denotes a significant difference with the control group ($P < 0.05$). Lower-case "b" denotes a significant difference with the OPO group ($P < 0.05$). Lower-case "c" denotes a significant difference with the OPO+MIX group ($P < 0.05$). WBC: white blood cell, LYM: lymphocyte, OPO: oxidized palm oil, MEN: mentha, THY: thymus, MIX; mixture of mentha and thymus.**

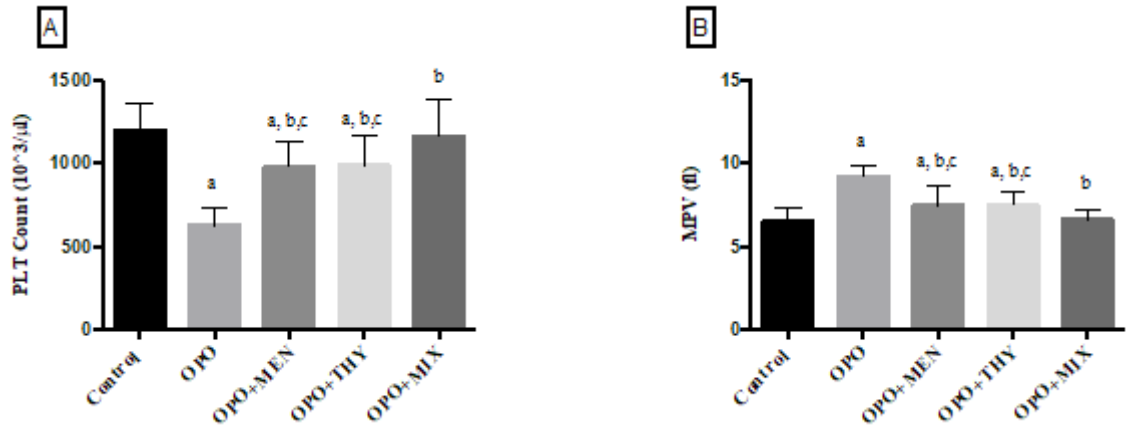

Fig. 3 **Effect of mentha and/or thymus extract on platelet indices [including (A) PLT count and (B) MPV] of rats fed a diet containing 15% OPO for 6 weeks. Data are presented as mean ± standard deviation (SD). Lower-case "a" denotes a significant difference with the control group ($P < 0.05$). Lower-case "b" denotes a significant difference with the OPO group ($P < 0.05$). Lower-case "c" denotes a significant difference with the OPO+MIX group ($P < 0.05$). PLT: platelet, MPV: mean platelet volume, OPO: oxidized palm oil, MEN: mentha, THY: thymus, MIX; mixture of mentha and thymus.**

Discussion

This study aimed to investigate the effect of *Mentha piperita* L and/or *Thymus vulgaris* in ameliorating the hematological changes induced by chronic consumption of oxidized palm oil in rats. Rats fed a diet containing OPO showed significant decreases in RBCs, Hct, MCV, Hb, MCH, MCHC, LYMs, and PLTs accompanied by significant increases in WBCs, RDW, and MPV compared to rats fed a normal diet (control group). These results agree with a previous study by Mesembe et al. (2004), who reported that chronic consumption of thermo-oxidized palm oil led to significant decreases in RBC count and Hb concentration and a significant increase in WBCs. They concluded that the consumption of thermo-oxidized palm oil results in anemia and leukocytosis in rats.[7] These results were further corroborated by Morshed et al. (2018), who found that long-term administration of heated palm oil to rabbits caused alterations in hematological indices characterized by decreased Hb concentrations and increased WBC counts.[21] Furthermore, Uddin et al. (2015) observed that Hb concentrations, Hct, and PLT counts were decreased while WBC count was increased in rabbits fed oxidized corn oil.[22] There are several possible explanations for these results. For example, disorders of bone marrow caused by free radicals contained in oxidized oil lead to alteration in the production of RBCs, WBCs, and PLTs. Moreover, the hazardous constituents of oxidized oils cause kidney damage, specifically partial tubular atrophy, and result in erythropoietin production failure which, in turn, causes a decrease in RBC count. The consumption of oxidized oils cause damage not only to bone marrow and kidneys but also to the liver.[23,24] It is likely, therefore, that damage to liver cells may have caused an insult in the body that contributed to the observed increase in WBC count as a normal physiologic response. Liver damage also causes a decrease in iron storage (such as ferritin and hemosiderin), which likely contributed to the decrease in Hb concentrations found in our

study. In addition, the decrease in Hb concentration could be a consequence of reduced uptake of iron by the damaged intestinal mucosa of the rats, which resulted in a reduced bioavailability of iron in the system.[4,7,25]

In contrast, rats fed a diet containing OPO and the aqueous extract of *Mentha piperita* showed significant increases in RBCs, Hct, MCV, Hb, MCH, MCHC, LYMs, and PLTs, accompanied by significant decreases in RDW, WBCs, and MPV compared to rats fed a diet containing OPO only. The aqueous extract of *Mentha piperita* leaves is rich in phytochemicals, especially phenols and flavonoids such as rosmarinic acid, chrysoeriol, caffeic acid, salvianic acid, luteolin, salvigenin, thymonin and carnosol.[26,27] Thus, the improvement in the hematological indices observed in our study could be attributed to the antioxidant activity of these compounds against free radicals.[9,23,24,28] To our knowledge, this is the first study to investigate the effect of *Mentha piperita* on hematological indices changes induced by chronic consumption of OPO. However, our findings are consistent with a previous study, which reported that treatment with aqueous extract of *Mentha piperita* leaves significantly enhanced the decrease in RBCs, Hb, MCH, MCHC, and PLTs in rats with ethanol-induced gastroduodenal ulcers compared to the untreated group.[11] Similar results were reported by Abdellatief et al. (2017), who found that treatment with peppermint essential oil rectified anemia resulting from diabetes and increased counts of leukocytes and PLTs in rats.[10] Moreover, Samarth et al. (2004) showed that *Mentha piperita* oil protected the hematopoietic system against lethal gamma radiation in rats.[9]

Not only *Mentha piperita* but also *Thymus vulgaris* treatment showed significant increases in RBCs, Hct, MCV, Hb, MCH, MCHC, LYMs and PLTs, accompanied by significant decreases in RDW, WBCs, and MPV compared to rats fed a diet containing OPO only. As with *Mentha piperita*, the aqueous extract of *Thymus vulgaris* leaves contains high amounts of phenolic compounds (including rosmarinic acid, quinic acid, luteolin-7-o-glucoside, protocatechuic acid, trans cinnamic acid, caffeic acid, naringenin, kaempferol, quercetin, apigenin, rutin, 3,4-di-O-caffeoylquinic acid, syringic acid, p-coumaric acid, naringin, acacetin, hyperoside (quercetin-3-o-galactoside), quercetrin (quercetin-3-o-rhamonosid), gallic acid, trans ferulic acid, apigenin-7-o-glucoside, 4-O-caffeoylquinic acid, chlorogenic acid, silymarin, catechin(+), luteolin and o-coumaric acid) which protect tissues and organs from oxidative damage induced by OPO.[12,29,30] No previous studies were found to reveal the protective effect of the aqueous extract of *Thymus vulgaris* on hematological indices in rats fed OPO. However, our results are in line with a study conducted recently by Abdallah et al. (2019), who found that administration of *Thymus vulgaris* to rats increased RBCs, Hb, MCHC, and LYMs reduced by the consumption of thermally oxidized sunflower oil.[12] Similarly, treatment with *Thymus vulgaris* extract increased Hct, MCV, Hb, MCH, MCHC, LYMs, and PLTs in lead-intoxicated rats.[31] Moreover, thymus oil was efficient in ameliorating the alteration in RBCs, Hct, Hb, and WBCs observed in rats exposed to γ-radiation.[14]

Our results also showed that treatment with the combination of *Mentha piperita* and *Thymus vulgaris* showed better improvement in the RBC, WBC, and PLT indices of rats when compared with rats fed OPO treated with either *Mentha piperita* or *Thymus vulgaris*. Furthermore, treatment with the mixture led to RBC, WBC, and PLT indices similar to those of rats fed a normal diet (control group). These results indicate that the mixture of the two plants was effective in rectifying the hematological changes induced by the consumption of OPO.

This research confirms previous findings and contributes to our understanding of the effects of *Mentha piperita* and *Thymus vulgaris* on hematological changes induced by OPO consumption. The administration of both plants separately was effective in improving hematological indices. However, the mixing of them showed better improvements and was effective in rectifying the hematological changes induced by OPO.

Ethics Approval

Animal procedures were performed with the approval (number 581-17) of the Ethics Committee of the King Fahad Medical Research Center and according to recommendations for the proper care and use of laboratory animals.

Author Contributions

M.A and N.N. conceived the idea and designed the study. M.A. carried out the experiments. H.A. and R.M. analyzed the data, performed the literature search and wrote the manuscript. N.N. reviewed the original manuscript. All authors have read and agreed to the published version of the manuscript.

Funding

This research received no external funding.

Conflicts of Interest

The authors declare no conflict of interest.

References

1. Falade AO, Oboh G and Okoh A I. Potential Health Implications of the Consumption of Thermally-Oxidized Cooking Oils - A Review. Polish J. Food Nutr. Sci. 2017;67(2):95–105.
2. Adedeji AA and Ngadi MO. 3-D Imaging of deep-fat fried chicken nuggets breading coating using X-ray micro-CT. Int. J. Food Eng. 2009;5(4):1–14.
3. Falade AO, Oboh G, Ademiluyi AO and Odubanjo OV. Consumption of thermally oxidized palm oil diets alters biochemical indices in rats. Beni-Suef Univ. j. Basic Appl. Sci. 2015;4(2):150–156.
4. Ani EJ, Nna VU, Obi CE and Udobong N J. Comparative Effects of Thermoxidized Palm Oil and Groundnut Oil Diets on some Haematological Parameters in Albino Wistar Rats. AJBAS 2015;9(5):181–184.
5. Oboh G, Falade AO and Ademiluyi AO. Effect of thermal oxidation on the physico-chemical properties, malondialdehyde and carotenoid contents of palm oil. Rivista Italiana Delle Sostanze Grasse 2014;91(1):59–65.
6. Ebong PE, Owu DU and Isong, EU. Influence of palm oil (Elaesis guineensis) on health. Plant Foods for Human Nutrition 1999;53(3):209–222.
7. Mesembe OE, Ibanga I and Osim EE. The Effects of Fresh and Thermoxided Palm Oil Diets on Some Haematological Indices in the Rat. Nigeria Journal of Physiological Scinces 2004;19(1): 86–91.
8. Johari NZ, Ismail IS, Sulaiman MR, Abas F and Shaari K. Acute toxicity and metabolomics analysis of hypocholesterolemic effect of *Mentha piperita* aqueous extract in Wistar rats. Int. J. Appl. Res. Nat. Prod 2015;8(1):1–11.

9. Samarth RM, Goyal PK and Kumar A. Protection of swiss albino mice against whole-body gamma irradiation by *Mentha piperita* (Linn.). Phytother. Res.: An International Journal Devoted to Pharmacological and Toxicological Evaluation of Natural Product Derivatives 2004;18(7):546–550.
10. Abdellatief SA, Beheiry RR and El-Mandrawy SAM. Peppermint essential oil alleviates hyperglycemia caused by streptozotocin- nicotinamide-induced type 2 diabetes in rats. Biomed. Pharmacother. 2017;95:990–999.
11. Zangeneh MM, Salmani S, Zangeneh A, Bahrami E and Almasi M. Antiulcer activity of aqueous extract of leaves of *Mentha piperita* in Wistar rats. Comp. Clin. Path. 2018;28(2):411–418.
12. Abdallah AA, Nasr El-Deen NA, Neamat-Allah AN and El-Aziz A, Heba I. Evaluation of the hematoprotective and hepato-renal protective effects of *Thymus vulgaris* aqueous extract on thermally oxidized oil-induced hematotoxicity and hepato-renal toxicity. Comp. Clin. Path. 2019;29(2):451–461.
13. Shittu OK, Musa F and Gbadamosi DF. Trypanocidal activity and heamatological changes in *T. brucei* infected rats treated with methanolic leaf extract of *Thymus vulgaris*. Int J Appl Biol Res 2013;5(1&2):109–114.
14. Nada AS. Modulating Efficacy of Thymus Oil in Rats Exposed to γ-Radiation. J. Radiat. Res. Appl. Sci. 2013;6(1):35–52.
15. Ng CY, Kamisah Y, Faizah O, Jubri Z, Qodriyah HMS and Jaarin K. Involvement of inflammation and adverse vascular remodelling in the blood pressure raising effect of repeatedly heated palm oil in rats. Int. J. Vasc. Med. 2012;2012:1–10.
16. Barbalho SM, Damasceno DC, Spada APM, Silva VSD, Martuchi KA, Oshiiwa M, et al. Metabolic profile of offspring from diabetic *Wistar* rats treated with *Mentha piperita* (peppermint). eCAM 2011, 2011.
17. Shati AA and Elsaid FG. Effects of water extracts of Thymus (*Thymus vulgaris*) and ginger (*Zingiber officinale* Roscoe) on alcohol abuse. FCT. 2009;47(8):1945–1949.
18. Nazri KAA, Fauzi NM, Buang F, Saad QHM, Husain K, Jantan I, et al. procumbens Standardised Extract Reduces Cholesterol Levels and Modulates Oxidative Status in Postmenopausal Rats Fed with Cholesterol Diet Enriched with Repeatedly Heated Palm Oil. ECAM. 2019;2019:1–15.
19. Ribeiro SC, Malheiros DF, Guilozki IC, Majolo C, Chaves FCM, Chagas, et al. Antioxidants effects and resistance against pathogens of Colossoma macrG Ilmum (Serassalmidae) fed *Mentha piperita* essential oil. Aquac. 2018;490:29–34.
20. Siti HN, Kamisah Y, Mohamed S and Jaarin, K. Effects of citrus leaf extract on aortic vascular reactivity in hypertensive rats fed repeatedly heated vegetable oil. Appl Physiol Nutr Metab. 2019;44(4):373–380.
21. Morshed MH, Ahmad MR, Rahim MA, Yeasmin F, Roy AK and Ibrahim M. Effect of long time heated palm oil on physico-chemical properties and pharmacology of rabbit. J. Eng. Sci. 2018;9(1):85–96.
22. Uddin I, Ahmad S, Haq IU, Nawab S, Uddin M and Nabi G. Thermally oxidized corn oil adversely affects serum biochemistry, blood hematology and liver histopathology of rabbits. Eur. J. Acad. Res. 2015;3(2):2362–2378.
23. Osman NN, Balamash KSA and Aljedaani MS. Impact of peppermint and thyme in ameliorating cardiac and hepatic disorders induced by feeding rats repeatedly heated fried oil. Int J Pharm Phytopharmacological Res 2019; 9(6):10–20.
24. Osman NN, Alsharari MA and Alsufiani HM. Peppermint (*Mentha piperita* L.) and Thyme (*Thymus vulgaris*) attenuate the Immune and Inflammatory Disorders in Rats Consumed Repeatedly heated Palm oil. Inter J Pharmaceutical Phytopharmacological Res. 2020;10:59–66.
25. Venkata RP and Subramanyam R. Evaluation of the deleterious health effects of consumption of repeatedly heated vegetable oil. Toxicol. Rep. 2016;3:636–643.
26. Ali J, Khan FA, Abbas S, Afridi MS and Rehman S. Comparative Phytochemical Evaluation of *Mentha piperita* Varieties (citrate and perpeta) Leaves and Stem Extracts. Spe. J. Biol. Sci 2017;3(3):1–5.
27. Brown N, John JA and Shahidi F. Polyphenol composition and antioxidant potential of mint leaves. Food Prod. Process. Nutr. 2019;1(1):1–14.
28. Patil SR, Patil RS and Godghate A. *Mentha piperita* Linn: Phytochemical, antibacterial and dipterian adulticidal approach. Int. J. Pharm. Pharm. Sci 2016;8(3):352–355.
29. Abobaker DM, Edrah SM, Jarood S, Alnade H and Kollab WA. Phytochemical Examination and Antibacterial Activities of Thyme Leaves and their Use with Goat's Milk as a Traditional Remedy. Acta Sci. Med. Sci. 2022;6:259–269.
30. Rtibi K, Selmi S, Wannes D, Jridi M, Marzouki L and Sebai H. The potential of *Thymus vulgaris* aqueous extract to protect against delayed gastric emptying and colonic constipation in rats. RSC Adva. 2019;9(36):20593–20602.
31. El-Boshy ME, Refaat B, Qasem AH, Khan A, Ghaith M, Almasmoum H, et al. The remedial effect of *Thymus vulgaris* extract against lead toxicity-induced oxidative stress, hepatorenal damage, immunosuppression, and hematological disorders in rats. Environ. Sci. Pollut. Res. 2019;26(22):22736–22746.