ORIGINAL ARTICLE

Evaluation of phytochemical constituents, antidotal and protective properties of *elaeis oleifera* fruit extract in oral organophosphate poisoning in Wistar rats

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ABSTRACT

Objectives: This study aimed at determining the phytochemical constituents and curative-protective effects of extract of *Elaeis oleifera* against oral organophosphate poisoning in rats. Methods: The oil of yellow palm fruits was extracted; phytochemical screening and anti-poisoning activity of the palm oil were determined using standard methods. Data were analysed using one-way ANOVA, Welch F, Tukey's pairwise and multiple comparison tests ($p \leq 0.05$). Major findings: Yellow palm oil contained flavonoids, alkaloids, carotenoid, steroids, terpenoids, saponins, tocopherol and phenols. The biochemical parameters (alanine aminotransferase (157.60±5.79 - 241.80±17.16IU/L), aspartate aminotransferase (192.80±8.25 - 221.20±22.29IU/L), gamma glutamyl transpeptidase (2.80±0.47 - 4.80±0.19g/dL) and total bilirubin (0.00±0.00 - 0.06±0.02mg/dL) of yellow palm oil treated groups of rats (A, B, C and D) showed significant recovery from oral organophosphate induced toxicities as against group E (566.90±19.49IU/L, 824.20±35.71IU/L, 653.60±70.11IU/L, 8.50±2.47g/dL, 0.08±0.05mg/dL) which received only poison with no reversal of toxicities, although with significant differences ($p = 0.0001$-0.0013; multiple comparisons). The haematological parameters (WBC (8.4±0.537x109/L, 7.2±0.769x109/L), RBC (8.4±0.328x1012/L, 8.0±0.313x1012/L), HGB (15.9±0.505g/dL, 15.4±0.487g/dL), HCT (48.4±1.533%, 46.3±1.592%), MCV (57.9±0.869fL, 58.8±1.099fL), MCH

(19.6±0.407pg, 19.9±0.367pg), MCHC (33.9±0.276g/dL, 34.1±0.304g/dL), PLT (555.4±80.223x109/L, 601.2±35.157x109/L), and MPV (7.46±0.385fL, 6.86±0.150fL)) of yellow palm oil treated groups of rats (B and D) were within normal ranges when compared to control group F (ascorbic acid treated, p = 0.0082-0.0250; multiple comparisons).
Conclusions: Yellow palm fruit oil contains phytochemicals which ameliorate symptoms and reverse oral dichlorvos induced biochemical and haematological toxicities in rats; suggesting its use in traditional medicine as anti-poison.

Keywords: Phytochemical, antidotal, preventive, *Elaeis oleifera*, oral, organophosphate poisoned rats.

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BACKGROUND

Acute organophosphate poisoning (OP) is a significant global health issue, with an estimated 3 million cases and over 220,000 deaths annually [1, 2]. These poisonings are often a result of intentional self-harm or accidental exposure to pesticides like chlorpyrifos and dimethoate, as well as highly volatile nerve agents like sarin and soman [3]. OPs disrupt neurotransmission by irreversibly inhibiting the enzyme acetylcholinesterase, which leads to the accumulation of acetylcholine at neuromuscular junctions, autonomic, and central nervous systems [4]. This overstimulation results in a cholinergic crisis, characterized by muscle weakness, fasciculations, seizures, coma, and potentially fatal respiratory failure [5].

The prognosis of OP poisoning is often unpredictable due to the difficulty in identifying the specific agent, dose, and half-life at the time of admission [6]. OP poisoning also leads to the production of reactive oxygen species (ROS), causing oxidative stress and lipid peroxidation in cell membranes [7]. The body attempts to counteract this damage using enzymatic antioxidants like superoxide dismutase (SOD), glutathione peroxidase (GPX), and catalase (CAT), as well as non-enzymatic antioxidants such as vitamins C and E, glutathione (GSH), and β-carotene [8].

In advanced nations, OP poisoning is typically treated with conventional pharmaceutical drugs like atropine. However, in developing countries, access to these treatments is challenging due to cost, lack of resources, and a shortage of trained personnel [9]. This contributes to a high global mortality rate of 20-70% [10, 11]. Consequently, many rural communities, who are most vulnerable to poisoning, often resort to more accessible and affordable traditional medicines [12].

In Nigeria and other parts of Africa, palm oil from *Elaeis guineensis* (red palm) and *Elaeis oleifera* (yellow palm) is a common folk remedy for poisoning, where their other products like kernel oil are common antidotes in each household and from where the two main species of oil palm trees (*Elaeis guineensis* and *Elaeis oleifera*) with three major varieties such as *dura, tenera* and *pisifera* are believed to have originated and later intentionally spread globally to other regions like the Americas and Asia because of its commercial viability [13]. The use of palm oil as an antidote is supported by studies confirming the anti-poisoning properties of its key constituents, such as vitamins E and A (tocopherols and tocotrienols), carotene, catechin, and ferulic acid [14, 15]. Additionally, flavonoids like quercetin, naringin, and rutin have been shown to protect against pesticide-induced toxicity through their antioxidant, anti-inflammatory, and anti-apoptotic activities [16, 17, 18]. This study, therefore, aimed to determine the phytochemical constituents, as well as the prophylactic and curative effects, of yellow palm fruit oil against oral organophosphate poisoning in rats.

METHODS

Aim, design and setting of the study

The aim of the study was to determine the phytochemical constituents and anti-poisoning and protective effects of the extract of *Elaeis oleifera* (yellow palm) fruit oil against organophosphate poisoning in albino Wistar rats. This study was an experimental study involving 35 rats divided into seven groups of five rats each (n = 5). The 35 rats were selected randomly from the rats raised for this study. The experiments were carried out in the Pharmacology Laboratory of the Kampala International University, Western Campus, Ishaka, Bushenyi District, Uganda using the anti-poisoning study procedures of Emna et al [19].

Characteristics of animals and description of materials

Healthy, mature male and non-pregnant female albino Wistar rats weighing 180 g and above were included in this study. The female rats were confirmed non-pregnant by separating them

from the male rats before they mature at 5-12 weeks in order to prevent mating and the female rats were checked for presence or absence of removal of hairs around their nipples and expansion of the specialised epidermis of their nipples or absence of expansion of the epidermis of their nipples to indicate pregnancy or non-pregnancy respectively; in case of very early maturity of the rats at 4 weeks of age [20]. Additionally, the female rats were observed for a marked reduction in voluntary physical activity and they were ascertained to possess the same high level of voluntary physical activity from the day of their separation from the males to the point of final determination of their pregnancy status [21]. Confirmation of presence or absence of pregnancy in the female rats was corroborated by monitoring the body weights of the female rats individually from the day of separation from the males up to the 14th to the 19th day and no significant weight variation was observed between the day of separation from the males and the 14th to the 19th day after separation when it was supposed that if the female rats were pregnant, their body weights would increase significantly from the day of separation to the 14th to 19th day [22].

Two main species of oil palm abound which include *Elaeis guineensis* (red palm fruit) and *Elaeis oleifera* (yellow palm fruit) and their oil is usually extracted from the fibrous mesocarp and stony palm kernel. The yellow palm fruit was selected for this study because the local population in South-Eastern and South-Southern regions of Nigeria have been using the oil from yellow palm fruits in preference to the oil from red palm fruits in the treatment of poisoning and other illnesses. The yellow palm fruits were picked in South-Eastern Nigeria from Aguluezechukwu and Ekwulobia communities in Aguata Local Government Area of Anambra State, where the use of this oil as anti-poison is extensive. The voucher specimen of the yellow palm fruit used in this study was labelled as EZE JOSEPH/KIU/22/001 and deposited in the Herbarium in the Pharmacognosy Laboratory of Kampala International University, Western Campus, Ishaka, Bushenyi, Uganda. The chemicals, reagents and equipment for phytochemical screening listed below were provided by the School of Pharmacy Laboratory, Kampala International University Western, Campus (KIU-WC), Ishaka, Bushenyi, Uganda. The chemicals included dilute ammonia solution, concentrated sulphuric acid, 5% ferric chloride solution, 0.1% ferric chloride solution, 1% aqueous hydrochloric acid, Mayer's reagent, sodium hydroxide solution, dilute hydrochloric acid, 2% aqueous hydrochloric acid, acetic anhydride and chloroform. The glassware included universal bottles (LMG01, Lab Manchester, UK), beakers (© Japson, Hi-Techsoft Services, India), and conical flasks (Superior Marienfeld, Germany) while the equipment were feeding needles (Cadence Science™ 7913, 18G, 01-290-10B, USA), aluminum cooking pot (locally manufactured along Basajjabalaba Road, Ishaka, Bushenyi, Uganda), dissecting kits (Royal 118 Dissecting set, Star Labs, Haryana, India), oven (Hot Air Sterilizer, 202-0, 0814002, Italy), water bath (Thermostat Water Bath, HH-4 (XMTD-205c, China), Weighing balance/scale (Electronic Scale, Model 30002, max 3kg, China) and stirrer (78-1 Magnetic Stirrer Hot Plate, 2400rpm, China).

Extraction of palm oil

The ripe yellow palm fruits were initially washed with clean water and parboiled in water for ten minutes and dried under shade in the laboratory for preservation till when required for use. The dried palm fruits were softened by boiling in water for 2 hours and the pulp was then immersed in warm water with thorough stirring. The oil was separated from the fibre and seeds into an aluminum cooking pot by first sieving with a basket and then with the smallest sized sieve (Plastic Sieve, Dacongming, UBuy, Uganda). The oily filtrate in the aluminum cooking pot was boiled on a hot plate (Digital Hot Plate, DB-2A, China) for five hours and

allowed to cool to enable scooping of the palm oil which set on top of the aqueous portion of the boiled filtrate into a fresh container. Traces of water in the oil were removed in a bout of gentle heating and concentration for ten minutes.

Phytochemical screening tests

Standard methods of Sofowora [23], Trease and Evans [24], and Harborne [25] were used to conduct chemical tests for preliminary qualitative phytochemical screening of yellow palm fruit oil for the presence of flavonoids, phenols, tannins, phlobatannins, saponins, terpenoids, alkaloids, steroids, carotenoid and tocopherols in test tubes (Superior Marienfeld, Germany).

Laboratory animal acquisition and maintenance

Male and female Wistar rats weighing 180 g and above were acquired, bred and housed in the Animal Facility Centre of the School of Pharmacy, KIU-WC. The animals were made to acclimatise within 14 days by separating them in a cage in a new partition of the animal house, supported with wood flakes, maintained at room temperature with sufficient aeration and environment naturally illuminated with 12 h of light and 12 h of darkness. The animals were made to acclimatise for a long period of 14 days because they were separated into a new partition of the same animal house and also in order to allow enough time for observation and confirmation of presence or absence of pregnancy. They were fed on standard diet (Nuvita® Animal Feed Ltd, Jinja Uganda) and permitted access to clean drinking water ad libitum. The animals used in the studies were subjected to humane treatment as ethically required according to the National Institute of Health Guide for the care and use of laboratory animals [26] and ethical guidelines for investigation of experimental pain in conscious animals [27]. Ethical approval to collect data using the rats in this study was granted by the Kampala International University School of Pharmacy Research and Ethics Committee (KIU-REC/2022/SP/020).

Anti-poisoning studies

The anti-poisoning activity of the yellow palm fruit extract (oil) was carried out according to the method described by Emna et al [19]. The animals were categorised into 7 treatment groups with 5 rats each (n = 5). Group A - was given dichlorvos (organophosphate) poison for one week and yellow palm oil for another one week, Group B - was administered with yellow palm oil first, and dichlorvos 30 minutes later. Group C - was given dichlorvos first, and yellow palm oil 30 minutes later, Group D - was administered with dichlorvos and yellow palm oil simultaneously. Group E - was given dichlorvos alone for two weeks. Group F - was given dichlorvos first and vitamin C (ascorbic acid) 30 minutes later (positive control) for two weeks and Group G - was given distilled water (negative control) for the same period. The toxic, non-lethal oral dose of the organophosphate administered through gavage to the rats was equivalent to a twentieth of the LD50 (25 mg/kg) which Hagar and Fahmy [28] reported to have been previously used by other investigators. A 0.01 g amount of the poison was diluted in 100 mL of distilled water following the methods of AOAC [29] and WHO [30]. Four (4) mL/kg of body weight of the oil extract was the volume of extract administered to the rats daily. Ascorbic acid was given as a positive control at a dose of 100 mg/kg for protection against the dichlorvos induced toxicity and oxidative stress [31]. After oral treatment of the rats with the palm oil and poison on the last day, the rats were sacrificed using cervical dislocation in their respective groups; and the blood of each rat was collected via cardiac puncture into plain vacutainer tubes and allowed to clot within 30 minutes and centrifuged at 3,000 revolutions per minute for 10 minutes to obtain serum for biochemical analysis and another set of whole blood was collected into heparinised vacutainer tubes ready for haematological analysis.

Biochemical tests

The resultant serum extracted from the clotted blood was subjected to biochemical enzymes assays to determine the protective and curative properties of the yellow palm oil against organophosphate poisoning. Commercial test kits used were obtained from appropriate sources and used for the assay of biochemical enzymes such as alanine aminotransferase (ALT), aspartate aminotransferase (AST)), alkaline phosphatase (ALP), gamma glutamyl transpeptidase (GGT) and total bilirubin (Bil T) in the serum at Kampala International University Teaching Hospital Clinical Chemistry Laboratory using the Humalyzer 2000, German model [32, 33]. The methods for determining ALT, AST, and ALP followed standard procedures [34, 35].

Haematological tests

The haematological assays were done to determine the protective property of the yellow palm oil extract using standard methods [32]. Commercial test kits used for the analysis were obtained from appropriate sources and used to measure all haematological parameters at Kampala International University Teaching Hospital Haematology and Blood Transfusion Laboratory. The unclotted whole blood samples in heparinized vacutainer tubes were used to estimate the complete blood count consisting of tests such as haemoglobin (Hgb), haematocrit (Hct or PCV), white blood cells counts (WBCs), red blood cells (RBCs) counts, and red cell indices such as mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC); platelets counts (PLT) and mean platelets volume (MPV) values using the automated haematology system analyzer (ADVIA 60 open tube; Bayer Corporation, Tarrytown, New York, USA).

Data analysis Data were analysed using Graph pad prism software (version 7.0) and past software and the major tools used were one-way ANOVA, Welch F, Tukey's pairwise and multiple comparison tests at $p \leq 0.05$ level of significance.

RESULTS

Phytochemical analysis

The results of the phytochemical analysis showed that *Elaies oleifera* (yellow palm fruit oil) extract contained flavonoids, phenols, saponins, terpenoids, alkaloids, steroids, carotenoids and tocopherols but no tannins and phlobatannins.

Biochemical assay

The results of the biochemical assay of the effects of *Elaeis oleifera* crude extract (yellow palm fruit oil) on oral dichlorvos-poisoned albino rats are shown in table 1. The results showed that group E (which received dichlorvos poison for two weeks) recorded the highest mean values of alanine aminotransferase (ALT, 566.90±19.49 IU/L), followed by group D (which received poison and palm oil simultaneously for two weeks, 241.80±17.16 IU/L), then group B (which received palm oil first and poison 30 minutes later for two weeks, 240.00±10.16 IU/L), group G (negative control, which received distilled water for two weeks, 233.70±14.71 IU/L), then group A (which received poison first week and palm oil second week, 192.80±7.67 IU/L), group C (which received poison first and palm oil 30 minutes later for two weeks, 188.20±3.70 IU/L) and then the lowest value was recorded by group F (positive control, which received poison first and ascorbic acid 30 minutes later for two weeks, 157.60±5.79 IU/L). The ALT results of the treated groups (C, A, B and D), appearing in ascending order as arranged, indicated statistically significant recovery of liver cells from the poison effect showing approximately the same range of ALT values when compared with groups F (positive control) and G (negative control) in contrast to the outrageously high

level of ALT for group E (received only poison for two weeks) (p = 0.0001 < 0.05 (Table 1). That means, group C recovered better than group A, then group B and then finally group D. The highest level of aspartate aminotransferase (AST) was observed in group E (which received only poison for two weeks, 824.20±35.71 IU/L). The next highest in rank was group C (received poison first and palm oil 30 minutes later for two weeks, 393.20±8.81 IU/L), then group D (received poison and palm oil simultaneously for two weeks, 221.20±22.29 IU/L), group G (negative control, received only distilled water for two weeks, 214.90±8.88 IU/L), group B (received palm oil and poison 30 minutes later for two weeks, 207.98±12.69IU/L), group F (positive control, received poison and ascorbic acid 30 minutes later for two weeks, 195.50±4.63 IU/L) and then group A (received poison first week and palm oil second week, 192.80±8.25 IU/L) which had the lowest value of AST. The treated groups A, B, D and C, arranged in ascending order of poison effect as displayed in table 1 were confirmed to be within the same statistically significant range (p value of 0.0001) when compared with group G (negative control), F (positive control) and group E that received only poison for two weeks at $p \leq 0.05$, indicating different levels of recovery from the effect of dichlorvos poisoning while group E (poison only for two weeks) showed a substantial increase in AST indicating no reversal of dichlorvos induced toxicity. That means, group A recovered better than group B, then group D and then finally group C.

The alkaline phosphatase (ALP) results pointed out that the highest value was seen in group E (poison only for two weeks, 653.60±70.11 IU/L). The next was group B (palm oil first and poison 30 minutes later for two weeks, 566.40±41.79 IU/L) trailed by group D (received poison and palm oil simultaneously for two weeks, 541.30±83.71 IU/L), A (poison first week and palm oil second week, 540.80±81.08 IU/L), F (positive control, poison first and ascorbic acid 30 minutes later, 467.70±33.37 IU/L), G (negative control, distilled water for two weeks, 432.20±71.02 IU/L) and then the lowest value was attained by group C (poison first and palm oil 30 minutes later for two weeks, 377.40±21.62 IU/L). The ALP results shown in Table 1 indicated the positively increasing recovery and protective effects of palm oil on the treated groups C, A, D and B which invariably exemplified different levels of statistically significant recovery and protection from dichlorvos induced poisoning (p = 0.00707 < 0.05) when compared with the groups F (positive control), G (negative control), group E (poison only for two weeks) which demonstrated no notable reversal of the dichlorvos induced toxicity.

Group E of rats (poison only for two weeks) exhibited the highest level of gamma glutamyl transpeptidase (GGT, 8.50±2.47 g/dL). The next in line was group G (negative control, distilled water for two weeks, 8.20±1.45 g/dL), trailed by group C (poison first and palm oil 30 minutes later for two weeks, 5.30±0.77 g/dL), D (simultaneous poison and palm oil for two weeks, 4.80±0.19 g/dL), F (positive control, poison first and ascorbic acid 30 minutes later for two weeks, 4.00±0.47 g/dL), A (poison first week and palm oil second week, 3.20±1.22 g/dL) and then group B (palm oil first and poison 30 minutes later for two weeks, 2.80±0.47 g/dL), which recorded the lowest value of GGT. Hence, there were different levels of statistically significant recovery from dichlorvos-induced poisoning as shown in Table 1 by the GGT values of the treated groups B, A, D and C, arranged in ascending order (p = 0.0129 < 0.05, multiple comparisons test) when compared with groups G (negative control), F (positive control) and group E (poison only for two weeks). That means, group B recovered better than group A, then group D and then finally group C.

Group E (poison only for two weeks) scored the highest level of total bilirubin (BilT) (0.08±0.05 mg/dL) among the other groups B (palm oil first

and poison 30 minutes later for two weeks, 0.06±0.02 mg/dL), G (negative control, distilled water for two weeks, 0.06±0.02 mg/dL), C (poison first and palm oil 30 minutes later for two weeks, 0.04±0.02 mg/dL), F (positive control, poison and ascorbic acid 30 minutes later for two weeks, 0.04±0.02 mg/dL), D (simultaneous poison and palm oil for two weeks, 0.02±0.02 mg/dL) and then A (poison first week and palm oil second week, 0.00±0.00 mg/dL) in that descending order. Statistical comparison with groups G (negative control), F (positive control) and E (poison only for two weeks) showed different degrees of statistically significant recovery from dichlorvos induced toxicity in terms of BilT secretion in the treated groups, with A having the best recovery, followed by D, C and B (p = 0.0001 < 0.05, multiple comparisons test).

In summary, the biochemical parameters (alanine aminotransferase, aspartate aminotransferase, alkaline phosphatase, gamma glutamyl transpeptidase and total bilirubin) of palm oil treated groups of rats (A, B, C and D) showed significant recovery from oral organophosphate induced toxicities as against group E which received only poison with no reversal of toxicities. There was no significant difference when palm oil treated groups were compared with groups F (ascorbic acid treated; positive control) p = 0.9998 (Welch F test) at $p \leq 0.05$ and when the palm oil pre-treated group B and post-treated group C were compared (p = 0.1133 (Tukey's pairwise comparison) at $p \leq 0.05$.

Haematological assay

In Table 2, group E which received poison only for two weeks (12.5±0.632x10^9/L) exhibited the highest level of white blood cells counts (WBC) in relation to groups A (poisoned first week and palm oil second week, 8.4±1.072x10^9/L), B (palm oil and poison 30 minutes later for two weeks, 8.4±0.537x10^9/L), F (positive control, poison first and ascorbic acid 30 minutes later for two weeks, 8.1±0.879x10^9/L), G (negative control, distilled water for two weeks, 7.4±1.666x10^9/L), D (simultaneously received poison and palm oil for two weeks, 7.2±0.769x10^9/L) and then group C (poison first and palm oil 30 minutes later for two weeks, 5.3±0.664x10^9/L) in which the lowest value of WBC was observed. Comparatively, the WBC counts of the treated groups C, D, B, and A as arranged in ascending order of protective and defensive response against dichlorvos induced poisoning is shown in Table 2. When the tested groups were compared statistically with group F (positive control), G (negative control) and group E (received poison only for two weeks), they were seen to fall within similar range of statistical significance indicating different shades of pronounced protective and defensive responses from the immune system to the poisoning induced by dichlorvos (p = 0.0011 < 0.05, multiple comparisons). That means A had the best protective and defensive response followed by B which recorded better response than D and D better than C.

Group B (which received palm oil and then poison 30 minutes later for two weeks, 8.4±0.328X10^{12}/L) topped the groups with the highest counts of red blood cells (RBC). Group B was followed in succession by other test groups E (8.1±0.269X10^{12}/L), D (8.0±0.313X10^{12}/L), A (8.0±0.266X10^{12}/L) and C (7.9±0.222X10^{12}/L), and trailed by control groups G (negative control, 7.8±0.108X10^{12}/L) and F (positive control, 7.2±0.202X10^{12}/L). The RBC counts of treated groups B, D, A, and C as shown in Table 2, and arranged here in descending order of response to the oxidative stress occasioned by the organophosphate poisoning were found to be statistically non-significant and also exhibiting statistically significant differences in their individual responses when compared with groups G (negative control), group F (positive control) and untreated group E (p = 0.0954 > 0.05, multiple comparisons); indicating the ready adaptation of the poisoned rats towards the

oxidative stress induced by dichlorvos. In summary, treated group B adapted to the dichlorvos induced toxicity better than group D, which showed better adaptation than A, and A better than C.

Group E which received poison only for two weeks responded to the poison with highest value of haemoglobin (HGB, 16.4±0.298 g/dL). The values of haemoglobin for rat groups B (15.9±0.505 g/dL), A (15.8±0.515 g/dL), D (15.4±0.487 g/dL), C (15.4±0.246 g/dL), G (14.7±0.820 g/dL) and F (14.6±0.436 g/dL) which followed respectively in a descending order of response to oxidative stress orchestrated by dichlorvos induced poisoning showed that test group B adapted to the oxidative stress better than group A, A better than group D and D better than group; when compared statistically with groups F (positive control), G (negative control) and untreated group E (poison for two weeks). But multiple comparisons of HBG values between groups showed no statistically significant responses ($p = 0.1779 > 0.05$).

Group E also recorded the highest level of haematocrit (HCT, 49.2±0.799%) as shown in Table 2, followed in a descending order by groups B (48.4±1.533%), A (46.9±1.484%), G (46.5±1.150%), D (46.3±1.592%), C (45.5±0.541%), and then F (44.3±1.115%); with groups A, C and D comparatively found within the same range as groups F (positive control) and G (negative control) while groups B values were found slightly increased when compared to the other treated and control groups testifying to the oxidative stress occasioned by dichlorvos. However, the responses among the groups were confirmed non-significant via multiple comparisons statistical tests results ($p = 0.2823 > 0.05$).

The mean cell volume (MCV) results in Table 2 indicated that the highest value was documented in group E (61.6±1.511 fL) which received poison only for two weeks. This trend was followed by groups F (60.1±0.849 fL), D (58.8±1.099 fL), G (58.7±0.815 fL), A (58.6±0.539 fL), and then by group C (57.4±0.898 fL) with the lowest MCV value. The MCV result indicated there were no significant responses by the treated and control groups as well as untreated group E (poison only for two weeks) when compared ($p = 0.0824 > 0.05$).

Group E recorded the highest value of mean cell haemoglobin (MCH, 20.6±0.366 pg). Group E was followed by groups F (20.2±0.219 pg), D (19.9±0.367 pg), B (19.6±0.407 pg), A (19.6±0.084 pg), C (19.5±0.298 pg), and then group G (19.3±0.389 pg). There were also no significant responses from the treated groups A, B, C, D, and the control groups F and G as well as untreated group E when ompared ($p = 0.8736 > 0.05$).

Group D had the highest values of mean cell haemoglobin concentration (MCHC, 34.1±0.304 g/dL), followed by groups B (33.9±0.276 g/dL), C (33.8±0.244 g/dL), E (33.6±0.237 g/dL), F (33.1±0.389 g/dL), and then group G (32.9±0.462 g/dL). No significant recoveries were recorded between the groups when compared with multiple comparison tests ($p = 0.1616 > 0.05$).

The PLT results shown in Table 2 revealed that the highest mean value of platelets counts was in group A ($660.6\pm28.719\times10^9$/L) which was followed by groups E ($640.0\pm43.229\times10^9$/L), F ($605.6\pm6.997\times10^9$/L, D ($601.2\pm35.157\times10^9$/L, B ($555.4\pm80.223\times10^9$/L, and trailed by group G ($390.2\pm64.145\times10^9$/L. The PLT results indicated that the values for treated groups B and D were within the same range as group F with statistically significant protection form the induced poisoning when groups A and C were compared to untreated group E ($p = 0.0082 > 0.05$ on multiple comparisons of PLT results.

Group G had the highest values of MPV, 8.14±0.437 fL among all the groups, followed by groups B (7.46±0.385 fL), F (7.22±0.132 fL), A (7.08±0.124 fL), C (7.06±0.150 fL), E (7.04±0.150 fL), and followed in succession. The MPV results showed that the treated groups A, B, C and D were within the same range of

statistical significance when controls F, G and the untreated group E were compared (p = 0.0250 < 0.05, multiple comparisons test). Aside from the above observations centred on the objectives of this study, diarrhoea was also observed in rats which were given only poison or poison first for a week before palm oil.

In summary, the haematological parameters (WBC, RBC, HGB, HCT, MCV, MCH, MCHC, PLT, and MPV) of yellow palm oil treated groups of rats (B and D) were within normal ranges when compared to control group F (ascorbic acid treated) except RBC for group B, MCHC for A, B, C, D; PLT for A and C and MPV for A, B, C and F which had higher values than the untreated group E. However, there were no significant differences between yellow palm oil treated groups A, B, C and D, and ascorbic acid treated group F (p = 0.9967; Welch F) at $p \leq 0.05$ and between the pre-treated group B and post-treated group C (p = 0.0839; Tukey's pairwise comparison at $p \leq 0.05$) when compared.

DISCUSSION

This study aimed to investigate the phytochemical composition and the antidotal and protective properties of *Elaeis oleifera* (yellow palm) fruit oil against oral organophosphate (dichlorvos) poisoning in Wistar rats. The findings confirm that yellow palm oil contains a variety of phytochemicals and demonstrates significant efficacy in reversing dichlorvos-induced biochemical and haematological toxicities.

Phytochemical Analysis

The phytochemical screening revealed the presence of flavonoids, alkaloids, carotenoids, steroids, terpenoids, saponins, tocopherols, and phenols in the yellow palm oil extract. The presence of these compounds is significant, as many of them are known for their antioxidant, anti-inflammatory, and protective properties [14, 15]. For instance, flavonoids and carotenoids have been shown to protect against pesticide-induced toxicity by mitigating oxidative stress and lipid peroxidation, which are common effects of organophosphate poisoning [7, 18]. This aligns with the traditional use of palm oil as a folk remedy for poisoning in regions like Nigeria [12, 16].

Biochemical analysis

The biochemical results showed that rats treated with yellow palm oil (groups A, B, C, and D) had a significant recovery from the elevated levels of alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), gamma glutamyl transpeptidase (GGT), and total bilirubin (BilT) that were observed in the poisoned, untreated group E. The high levels of these enzymes in group E indicate severe liver damage, a well-documented consequence of organophosphate toxicity [28, 36]. The ameliorative effects of the palm oil, particularly the recovery observed in groups B and C, demonstrate its curative and protective potential. This is further supported by the fact that the palm oil-treated groups showed similar biochemical parameter values to the positive control group F, which received ascorbic acid. This is particularly noteworthy as ascorbic acid (vitamin C) is a well-known antioxidant used to combat oxidative stress, a key pathological mechanism of organophosphate poisoning [31, 37].

Haematological analysis

Furthermore, the haematological parameters showed that the palm oil-treated groups adapted better to the oxidative stress induced by dichlorvos. The white blood cell (WBC) counts in the treated groups (A, B, C, and D) were significantly lower than in the untreated poisoned group (E), and within a normal range. This suggests that the palm oil reduced the inflammatory response and defensive reaction of the immune system to the poisoning [38]. Similarly, the red blood cell (RBC),

haemoglobin (HGB), and haematocrit (HCT) values, while showing varied responses, generally indicated a better adaptation to the oxidative stress in the treated groups compared to the untreated group, which is consistent with the protective role of the oil's phytochemicals [14, 15].

Conclusion

In conclusion, the findings of this study support the traditional use of yellow palm oil as an anti-poisoning remedy. The rich phytochemical content of the oil, particularly its antioxidant components, appears to be responsible for its ability to ameliorate and reverse the biochemical and haematological toxicities induced by oral organophosphate poisoning. Given the high mortality rate of organophosphate poisoning and the limited access to conventional treatments in developing countries, the use of *Elaeis oleifera* fruit oil presents a promising, accessible, and affordable alternative for its management.

Recommendations

The results of this study are a call for further research into the specific mechanisms of action and isolation of the active compounds responsible for the observed effects.
Histopathological studies should also be done to determine the existence, extent and repair of organ damage. Similar studies should be done on the kernel extract (oil) and other parts of the palm tree to further identify and isolate the active ingredients in this plant that are involved in the antioxidant activity.

List of abbreviations

A: Rat group which received poison 1st week and palm oil 2nd week.
B: Rat group which received palm oil first and poison 30 minutes later for two weeks.
C: Rat group which received poison first and palm oil 30 minutes later for two weeks.
CAT: Catalase.
D: Rat group which received poison and palm oil simultaneously for two weeks.
E: Rat group which received poison only for two weeks.
F: Rat group (positive control) which received poison first and ascorbic acid 30 minutes later for two weeks.
G: Rat group (negative control) which received distilled water for two weeks.
GPX: Gluthathione peroxidase.
HBG: Haemoglobin.
HCT: Haematocrit.
MCH: Mean Cell Haemoglobin.
MCHC: Mean Cell Haemoglobin Concentration.
MCV: Mean Cell Volume.
MPV: Mean Platelets Volume.
PLT: Platelets.
RBC: Red Blood Cells.
SEM: Standard Error of the Mean.
SOD: Superoxide dismutase.
WBC: White Blood Cells.

DECLARATIONS

Ethics approval

The studies involved use of animals and therefore was approved ethically by the Kampala International University School of Pharmacy Research and Ethics Committee (KIU-REC/2022/SP/020) based on the premise that the experiments would be conducted humanely and with less pain according to the National Institute of Health guide for the care and use of laboratory animals [26] and ethical guidelines for investigation of experimental pain in conscious animals [27].

Consent for publication

The consent of all the authors were solicited and secured to authorise this publication.

Availability of data and material

Data and materials would be made available when required.

Competing interests

The authors declare no competing interests.

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Authors' contributions

JOCE initiated the work. Authors JOCE, DM, AAA, COO, AGO, JKT, BEU, OED and EA designed the study, wrote and corrected the protocol. Authors JOCE, DM, AAA and JCE collected the data. Authors JOCE, DM, SOO, COO, BEU, OED, EA, PMA, OPCU and AGO wrote the protocol and the first draft of manuscript, searched for literature, analysed the data, read through the data and made corrections. Authors JOCE, DM, JCE, JKT, GJU and AAA managed the experimental processes, read through and made corrections to the manuscript draft. Authors COO, JCE, BEU, OED, SOO, JKT, AE, PMA, OPCU and AAA read through and made corrections to the manuscript draft. All authors read and approved the manuscript for publication.

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TABLES

Table 1: Anti-poisoning effects of yellow palm oil on oral dichlorvos-induced poisoning of biochemical profiles of rats.

Groups	Biochemical parameters of rats for yellow palm oil (mean ± SEM)				
	ALT (IU/L)	AST (IU/L)	ALP (IU/L)	GGT (g/dL)	Bil T (mg/dL)
A	192.80±7.67	192.80±8.25	540.80±81.08	3.20±1.22	0.00±0.00
B	240.00±10.16	207.98±12.69	566.40±41.79	2.80±0.47	0.06±0.02
C	188.20±3.70	393.20±8.81	377.40±21.62	5.30±0.77	0.04±0.02
D	241.80±17.16	221.20±22.29	541.30±83.71	4.80±0.19	0.02±0.02
E	566.90±19.49	824.20±35.71	653.60±70.11	8.50±2.47	0.08±0.05
F	157.60±5.79	195.50±4.63	467.70±33.37	4.00±0.47	0.04±0.02
G	233.70±14.71	214.90±8.88	432.20±71.02	8.20±1.45	0.06±0.02
p value	0.0001	0.0001	0.00707	0.0129	0.0001

Key: ALT: Alanine Aminotransferase; AST: Aspartate Aminotransferase; ALP: Alkaline Phosphatase; GGT: Gamma Glutamyl Transpeptidase; BilT: Total Bilirubin; SEM: Standard

Error of the Mean; Groups: A (poison first week and palm oil second week); B (palm oil first and poison 30 minutes later for two weeks); C (poison first and palm oil 30 minutes later for two weeks); D (simultaneous poison and palm oil); E (poison only for two weeks); F (positive control; poison first and ascorbic acid 30 minutes later for two weeks) and G (negative control, distilled water only for two weeks).

Table 2: Anti-poisoning effects of yellow palm oil on oral dichlorvos-induced poisoning of haematological profiles of rats.

Groups	Haematological parameters for yellow palm oil (mean ± SEM)								
	WBC ×10⁹/L	RBC ×10¹²/L	HGB (g/dL)	HCT (%)	MCV (fL)	MCH (pg)	MCHC (g/dL)	PLT ×10⁹/L	MPV (fL)
A	8.4±1.072	8.0±0.266	15.8±0.515	46.9±1.484	58.6±0.539	19.6±0.084	33.6±0.167	660.6±28.719	7.08±0.124
B	8.4±0.537	8.4±0.328	15.9±0.505	48.4±1.533	57.9±0.869	19.6±0.407	33.9±0.276	555.4±80.223	7.46±0.385
C	5.3±0.664	7.9±0.222	15.4±0.246	45.5±0.541	57.4±0.898	19.5±0.298	33.8±0.244	609.8±31.198	7.06±0.150
D	7.2±0.769	8.0±0.313	15.4±0.487	46.3±1.592	58.8±1.099	19.9±0.367	34.1±0.304	601.2±35.157	6.86±0.150
E	12.5±0.632	8.1±0.269	16.4±0.298	49.2±0.799	61.6±1.511	20.6±0.366	33.6±0.237	640.0±43.229	7.04±0.150
F	8.1±0.879	7.2±0.202	14.6±0.436	44.3±1.115	60.1±0.849	20.2±0.219	33.1±0.389	605.6±6.997	7.22±0.132
G	7.4±1.666	7.8±0.108	14.7±0.820	46.5±1.150	58.7±0.815	19.3±0.389	32.9±0.462	390.2±64.145	8.14±0.437
p value	0.0011	0.0954	0.1779	0.2823	0.0824	0.8736	0.1616	0.0082	0.0250

Key: WBC: White Blood Cells RBC: Red Blood Cells; HBG: Haemoglobin; HCT: Haematocrit; MCV: Mean Cell Volume; MCH: Mean Cell Haemoglobin; MCHC: Mean Cell Haemoglobin Concentration; PLT: Platelets; MPV: Mean Platelets Volume; SEM: Standard Error of the Mean; Groups: A (poison first week and palm oil second week); B (palm oil first and poison 30 minutes later for two weeks); C (poison first and palm oil 30 minutes later for two weeks); D (simultaneous poison and palm oil); E (poison only for two weeks); F (positive

control; poison and ascorbic acid 30 minutes later for two weeks) and G (negative control, distilled water only for two weeks).