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Differential Effect of Fresh and Dried *Jatropha tanjorensis* Leaves on Dichlorvos- Induced Renal Injury

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ABSTRACT

Background: *Dichlorvos (DDVP), an organophosphate insecticide, is associated with renal toxicity due to oxidative and cellular damage. Jatropha tanjorensis, a medicinal leafy vegetable widely consumed in West Africa, possesses antioxidant properties; however, the effect of leaf processing on its nephroprotective potential remains unclear. This study evaluated the differential effects of fresh and dried Jatropha tanjorensis leaves on DDVP-induced renal injury. A total of thirty male mice were assigned into six groups: control (A), DDVP only (B), fresh J. tanjorensis only (C), dried J. tanjorensis only (D), DDVP + fresh leaves (E), and DDVP + dried leaves (F). Serum creatinine, urea, total protein, albumin, and bilirubin were assessed, alongside histopathological evaluation of kidney tissues. Results show DDVP exposure caused significant renal dysfunction, evidenced by elevated serum creatinine, Albumin, Total Protein, Total Bilirubin and urea levels with severe histopathological alterations including tubular degeneration, epithelial vacuolation, tubular dilation, and interstitial expansion. Co-administration of fresh J. tanjorensis leaves significantly reduced creatinine and urea levels and partially preserved renal architecture compared with the DDVP-only group. In contrast, dried leaves offered limited protection, as biochemical and histological abnormalities persisted despite partial structural preservation.*

Keywords: *Dichlorvos, Jatropha tanjorensis, nephroprotective, kidney biomarkers, histopathology.*

1. Introduction

The increasing use of synthetic pesticides in agriculture and domestic pest control has raised serious public health and environmental concerns. Among these chemicals, dichlorvos (2,2- dichlorovinyl dimethyl phosphate; DDVP) is a widely used organophosphate insecticide due to its rapid insecticidal action and broad-spectrum efficacy. However, extensive evidence indicates that DDVP exposure poses significant toxicological risks to non-target organisms, including humans and experimental animals [1,2,3]Chronic or acute exposure to DDVP has been implicated in multi-organ toxicity, with the kidney being one of the most vulnerable organs.

The kidney plays a pivotal role in maintaining homeostasis through filtration of blood, excretion of metabolic wastes, and regulation of fluid and electrolyte balance. Due to its high blood perfusion rate and involvement in xenobiotic elimination, the kidney is particularly susceptible to toxic insults [4]. Renal dysfunction induced by organophosphate pesticides is commonly manifested by elevated serum creatinine and urea levels, indicating impaired glomerular filtration and tubular reabsorptive capacity [5, 6]. These functional impairments are often accompanied by structural abnormalities such as tubular epithelial degeneration, cytoplasmic vacuolation, lumen dilation, and interstitial expansion.

The pathogenesis of DDVP-induced renal injury has been largely attributed to oxidative stress and inflammation. DDVP metabolism generates excessive reactive oxygen species (ROS), leading to lipid peroxidation, protein oxidation, mitochondrial dysfunction, and cellular apoptosis within renal tissues [7]. Persistent oxidative damage disrupts renal architecture and compromises nephron integrity, ultimately resulting in functional decline. Histopathological examinations in experimental models consistently support these biochemical disturbances, highlighting the strong correlation between kidney biomarkers and structural damage [2].

In recent years, increasing attention has been directed toward the use of medicinal plants as potential therapeutic agents against chemically induced organ toxicity. Plant-derived products are rich sources of natural antioxidants and bioactive compounds that exhibit cytoprotective, and free radical scavenging properties [8]. Such properties make medicinal plants attractive candidates for ameliorating pesticide-induced renal injury, particularly in low-resource settings where exposure risk is high and access to conventional therapies may be limited.

Jatropha tanjorensis is a tropical leafy vegetable widely consumed and used medicinally in West Africa. Traditionally, it has been employed in the management of various ailments, including anemia, metabolic disorders, and inflammatory conditions [9]. Phytochemical analyses of *J. tanjorensis* leaves have revealed the presence of flavonoids, phenolic compounds, saponins, and alkaloids—constituents known for their

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antioxidant and organ-protective activities [9, 10]. Experimental studies have reported that *J. tanjorensis* exhibits strong free-radical scavenging ability and may modulate oxidative stress-related tissue damage.

Despite its widespread use, limited information is available on the renal safety and protective potential of *J. tanjorensis* under conditions of chemical toxicity. Additionally, plant processing methods, such as drying, are known to significantly influence the stability and bioavailability of phytochemicals. Drying may lead to degradation or loss of heat and air-sensitive compounds, thereby altering the biological activity of plant extracts. Therefore, the form in which medicinal plants are consumed—fresh or dried—may crucially determine their therapeutic efficacy or potential toxicity.

Few studies have comparatively evaluated the effects of fresh versus dried plant materials in mitigating pesticide-induced renal injury. Understanding these differential effects is important for optimizing plant-based therapeutic strategies and ensuring safety in traditional medicine practices.

2.0. Experimental location

The experiment was carried out in the animal house of the Department of Animal and Environmental Biology, Rivers State University, Port Harcourt. (Coordinates 4° 48'14''N 6° 59' 12''E).3.0 Objectives

1. To evaluate the effect of dichlorvos exposure on renal function by assessing serum creatinine, urea, albumin, total protein, and bilirubin levels in experimental animals.
2. To determine the effects of administering fresh and dried *Jatropha tanjorensis* leaves on kidney function and renal histoarchitecture.
3. To compare the renal protective effects of fresh versus dried *Jatropha tanjorensis*
leaves following dichlorvos-induced nephrotoxicity.
4. To correlate biochemical alterations in kidney markers with histopathological changes in renal tissues across all treatment groups.

4.0 Materials and Methods

4.1 Experimental animals and Management

Thirty sexually mature male mice (mean weight 22.45±3.05g) were used in the study. The mice were housed individually in cages under standard conditions. They were provided with clean water and rodent pellet *ad libitum*. All experiments were conducted according to the Institutional Animal Care Protocols at the Rivers State University, Port Harcourt, Nigeria and followed guidelines for the ethical treatment of experimental animals.

4.2 Experimental Design and Procedure

A total of thirty (30) Swiss matured male mice were assigned to six (6) groups (A - F) of five
(5) mice each. Group A served as the control. Group B received 5mg/kg/bw/day of DDVP only. Group C received 250mg/kg/bw of fresh *Jatropha tanjorensis leaves* extract. Group D received 5mg/kg/bw/day of DDVP and 250mg/kg/bw/day of dried *Jatropha tanjorensis leaves*. Group E received 5mg/kg/bw/day of DDVP and 250mg/kg/bw/day of fresh *Jatropha tanjorensis* extract. Group F received 5mg/kg/bw/day and 250mg/kg/bw/day of dried *Jatropha tanjorensis*. All the groups were exposed to their treatment by oral gavage for thirty - five (35) days.

4.3 Collection of Blood Samples

After the final treatment, the animals were fasted for 24 hours with free access to water. Blood samples were collected under isoflurane inhalation anesthesia by retro-orbital puncture using a heparinized capillary tube inserted into the medial canthus of the eye. Blood was collected into labelled plain tubes, allowed to clot, and centrifuged at 3000rpm for 10 minutes to separate serum for determination of kidney biomarkers such as Total Protein (TP), Creatinine (CR), Urea (UR), Albumin (ALB), and Total Bilirubin (TB). Total protein was analyzed using the spectrophotometric method of biuret, Bradford and erythrosine-b-b, Creatinine and ureawere done using the enzymatic method (Bradford 1976), Total bilurubin (TB) and creatinine were determined as reported in [11, 12]. Albumin (ALB) concentration was assayed according to Sigma Diagnostics based on the procedure of [13,14].

4.4 Histopathological analysis of the Kidney

For each mouse, 0.5g of the kidney was fixed in 10% v/v buffered formaldehyde and dehydrated through ascending grades of ethanol, cleared in xylene and embedded in parafin wax and sectioned with a digital microtome at 5µm thick. Histological sections mounted on slides were stained with Hematoxylin and Eosin (H&E). Photomicrographs were generated with a digital Microscope Biosphere Miller B with an image processor DN2-Microscopy Image Processing Software at X40 magnification.

4.5. Statistical analysis

The results were statistically analyzed and expressed as mean± standard error (SE). For comparison between groups, the one-way analysis of variance (ANOVA) and correlation of the JASP version 0.18.3 and the software used was Microsoft Excel 365.

5.0 Results

5.1 The effect of dichlorvos (DDVP) and *Jatropha tanjorensis* leaf treatments on renal biomarkers are presented in Table 5.1.

Table 5.1: Effect of DDVP and Jatropha tanjorensis leaf treatments on renal biomarkers

GRPS	CR	TB	ALB	TP	UR
A	22.6±0.93^{a}	7.82±0.25^{a}	33.60±2.25^{ab}	59.60±0.68^{b}	3.18±0.10^{a}
B	91.40±1.60^{d}	13.76±0.62^{c}	47.20±0.66^{d}	73.62±0.75^{c}	4.64±0.05^{c}
C	92.00±3.79^{d}	11.13±0.60^{b}	41.01±1.15^{cd}	61.33±1.20^{b}	4.83±0.07^{c}
D	81.60±0.93^{c}	8.64±0.22^{a}	33.03±1.14^{a}	51.60±1.08^{a}	3.66±0.06^{b}
E	73.33±2.33^{b}	10.67±0.27^{b}	40.02±0.58^{bc}	61.67±0.88^{b}	3.67±0.09^{b}
F	94.33±0.67^{d}	7.90±0.31^{a}	34.31±1.20^{abc}	53.67±1.45^{a}	4.63±0.03^{c}

**Values are expressed as mean ± SEM. Means with different superscript letters within the same column are significantly different ($p < 0.05$).*

Serum creatinine (CR) levels were significantly lower in the control group (Group A: 22.60 ± 0.93 g/dl). A pronounced increase in creatinine was observed in the DDVP-only group (Group B: 91.40 ± 1.60g/dl). Similarly, elevated creatinine concentrations were recorded in groups C and F, while group D showed a moderate increase. Group E exhibited a significantly reduced creatinine level (73.33 ± 2.33g/dl) compared with Group B.

Total bilirubin (TB) concentration was significantly highest in Group B (13.76 ± 0.62g/dl). Groups C and E showed intermediate values, whereas Groups A, D and F had comparatively lower and similar bilirubin levels.

Albumin (ALB) levels varied significantly across the groups. The highest albumin concentration was recorded in Group B (47.20 ± 0.66 g/dl), followed by groups C and E. Groups A (33.60 ± 2.25 g/dl) and D (33.03 ± 1.14 g/dl) recorded the lowest albumin values, while group F showed a moderate level.

Total protein (TP) levels were significantly elevated in Group B (73.62 ± 0.75g/dl). Intermediate total protein values were observed in Groups C and E, whereas Groups A and D recorded lower values. Group F showed a reduced total protein concentration.

Serum urea (UR) concentration was significantly lower in the control group (3.18 ± 0.10 g/dl). Higher urea levels were observed in groups B, C and F. Groups D (3.66 ± 0.06g/dl) and E (3.67 ± 0.09g/dl) showed moderate urea values.

5.2: Histopathological analysis of kidney tissue of mice exposed to DDVP and *Jatropha tanjorensis*

Fig 5.2a shows kidney tissue from group A (control) with slight tubular epithelial changes, a well-preserved glomerular structure with largely intact tubular organization. Glomeruli appear mostly intact with preserved capillary loops.

Fig 5.2b shows the kidney tissue of group B animals (DDVP only) marked with numerous tubular epithelial cells. Many tubules appeared dilated with loss of a clear tubular lumen. Swollen epithelial cells, vacuolated cytoplasm and widened interstitial area.

Fig 5.2c shows the photomicrograph of group C animals administered fresh *J. tanjorensis* leaves. Numerous elongated tubular structures are visible, dilated lumen, flattened epithelial lining, loss of clear cellular boundaries and marked tubular dilation (MTD).

Fig 5.2d shows group D animals administered dried *J. tanjorensis* leaves. The tissue architecture appeared distorted with tubular degeneration. Severe loss of normal renal architecture marked with interstitial expansion.

Fig 5.2e is the photomicrograph of group E animals administered DDVP and fresh *J. tanjorensis* leaves. The kidney tissue shows signs of tissue alteration, several glomeruli arevisible, degenerated epithelial lining, cytoplasmic vacuolation in some cells, interstitial areas appear expanded.

Fig 5.2f is the photomicrograph of group F animals administered DDVP and dried *J. tanjorensis* leaves showing numerous closely packed tubules, loss of distinct lumen, epithelial cells appear swollen (vacuolated cytoplasm), tubular degeneration, interstitial space appears widened with small scattered nuclei. However, the kidney structure is partially preserved.

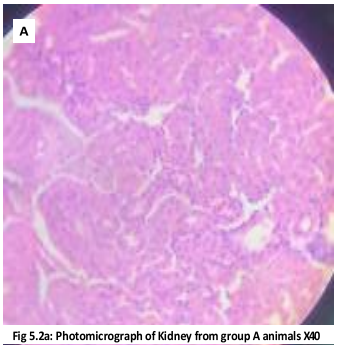

Fig 5.2a: Photomicrograph of Kidney from group A animals X40

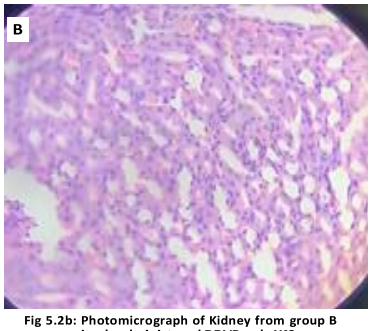

Fig 5.2b: Photomicrograph of Kidney from group B animals administered DDVP only X40

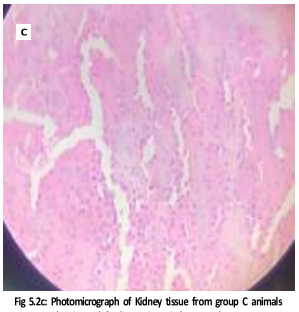

Fig 5.2c: Photomicrograph of Kidney tissue from group C animals administered *fresh J.tanjorensis* leaves only X40

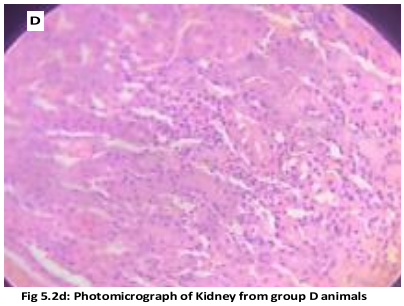

Fig 5.2d: Photomicrograph of Kidney from group D animals administered dried *J.tanjorensis* leaves only X40

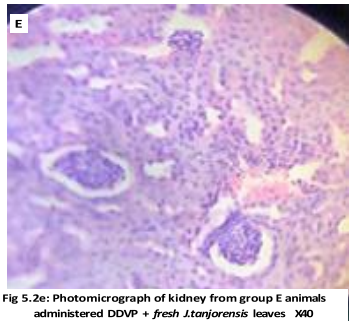

Fig 5.2e: Photomicrograph of kidney from group E animals administered DDVP + *fresh J.tanjorensis* leaves X40

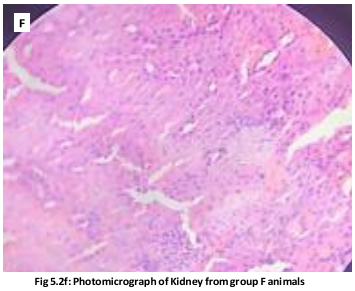

Fig 5.2f: Photomicrograph of Kidney from group F animals administered DDVP + dried *J.tanjorensis* leaves X40

6.0 Discussion

The present study demonstrates that exposure to dichlorvos (DDVP) induces marked renal dysfunction, as evidenced by significant elevations in serum creatinine and urea levels in the DDVP-only group (Group B) compared with the control. Elevated creatinine and urea are established indicators of impaired glomerular filtration and reduced renal excretory capacity [5,6]. These biochemical findings correlate strongly with the histopathological observations in Group B, which revealed tubular epithelial cell swelling, cytoplasmic vacuolation, tubular dilation, loss of clear tubular lumen, and widening of the interstitial spaces. Such lesions are characteristic features of pesticide-induced nephrotoxicity and are commonly associated with organophosphate exposure [2].

The severe renal structural damage observed following DDVP administration is consistent with reports that organophosphate pesticides generate excessive reactive oxygen species (ROS), leading to oxidative stress, lipid peroxidation, and subsequent cellular degeneration in renal tissues [7]. Tubular epithelial injury and interstitial expansion disrupt normal tubular reabsorption and glomerular filtration, thereby explaining the accumulation of nitrogenous waste products seen in this study.

Animals administered *Jatropha tanjorensis* alone (Groups C and D) exhibited renal structural alterations despite the absence of DDVP exposure.

In Group C, marked tubular dilation and epithelial flattening were evident, whereas Group D showed more pronounced architectural distortion with tubular degeneration and interstitial expansion. These observations suggest that although *J. tanjorensis* is widely regarded as medicinal, its renal effects may vary depending on preparation method, dosage, and phytochemical composition [10]. The relatively poor outcome in Group D (dried leaves only) further suggests that certain preparations of herbal products may themselves induce renal stress or lack benefit if active compounds are diminished. This observation resonates with prior warnings in the literature that some herbal medicines may pose nephrotoxic risk depending on preparation, dosage, and purity [15].

Previous studies have similarly cautioned that some herbal preparations may exert nephrotoxic effects or induce renal stress when bioactive compounds are improperly processed or concentrated [16].

Co-administration of DDVP with fresh *J. tanjorensis* leaves (Group E) significantly reduced serum creatinine and urea levels compared with the DDVP-only group, indicating partial restoration of renal function. This biochemical improvement was supported by histopathological findings, which showed relatively preserved glomerular structures and fewer degenerative changes, although some residual vacuolation and interstitial expansionpersisted. The observed nephroprotective effect is likely attributable to antioxidant and anti-inflammatory phytochemicals present in fresh leaves, which may counteract DDVP-induced oxidative stress and stabilize renal cell membranes [8, 10].

In contrast, co-administration of DDVP with dried *J. tanjorensis* leaves (Group F) did not markedly improve creatinine or urea levels relative to the DDVP-only group. Histological examination revealed persistent tubular degeneration, swollen epithelial cells with vacuolated cytoplasm, and widened interstitial spaces, although partial preservation of renal structure was still evident. The reduced protective efficacy of dried leaves may be due to degradation or loss of heat- or air-sensitive bioactive constituents during the drying process, which has been shown to significantly alter phytochemical potency [16].

The strong concordance between elevated biochemical indices of renal dysfunction (creatinine and urea) and the observed histopathological lesions confirms that DDVP consistently induces nephrotoxicity, while the protective influence of fresh *Jatropha tanjorensis* leaves is evident at both functional and structural levels. A recent study on *Jatropha dioica* showed that pre-treatment with its extract attenuated ischemia–reperfusion– induced renal injury in rats, with reductions in creatinine and oxidative stress markers, and partial restoration of histology [17].

These findings suggest a plausible mechanism: DDVP toxicity likely initiates oxidative stress, lipid peroxidation, and cellular damage, leading to tubular necrosis and glomerular dysfunction. The bioactive phytochemicals in fresh *J. tanjorensis* likely exerts antioxidant, anti-inflammatory, and membrane-stabilizing effects, thereby protecting renal cells and preserving structural integrity. The diminished effect seen with dried leaves supports the notion that processing (drying) may degrade such compounds, reducing their efficacy.

Conclusion

DDVP induces marked nephrotoxicity, while fresh *Jatropha tanjorensis* leaves provide partial protection against DDVP-induced renal injury. Drying appears to reduce the nephroprotective efficacy of the plant, highlighting the importance of processing methods in herbal therapeutics.

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