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Pyridoxal Phosphate protects against aluminium chloride-induced neurobehavioural and neuromorphological alterations in the rat cerebral cortex

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ABSTRACT

The increasing prevalence of neurodegenerative diseases has necessitated the search for affordable medicines for better management. Pyridoxal phosphate (PLP) has been reported to improve cognition in humans and rodents; however, its possible benefits in the management of neurodegenerative diseases like Alzheimer's disease has been scarcely examined. This study assessed the effects of PLP on aluminium Chloride-induced neurobehavioural and histomorphological changes in rat cerebral cortex. Forty male Wistar rats weighing 130-150 g each were distributed into four groups (n=10). Group A received distilled water (10 ml/kg), group B received $AlCl_3$ (50 mg/kg), group C and D received $AlCl_3$ with PLP at 100 mg/kg and 200 mg/kg respectively. Pyridoxal phosphate was administered by gavage on days 1-15 while $AlCl_3$ was administered orally on days 16-30. The results revealed a significant increase in horizontal locomotion, rearing and grooming in groups treated with PLP compared to $AlCl_3$. Spatial working memory increased significantly improved in groups treated with PLP compared to $AlCl_3$. An increase in MDA and inflammatory cytokines was observed with $AlCl_3$ compared to control with a reversal of these parameters with PLP supplementation. Results of histomorphological examination revealed evidence of neurodegeneration in the $AlCl_3$ groups, with PLP supplementation rendering some protection against neuronal injury. In conclusion this study revealed that PLP supplementation was protective against $AlCl_3$-induced neurobehavioural, oxidative stress, neuroinflammation and neuromorphological injury in rats. However, more studies are required to determine its benefits in humans.

KEYWORDS: Aluminium Chloride; Neurotoxicity; Neuroinflammation; Neuroprotection; Vitamin B6

1. Introduction

Neurodegenerative diseases (NDDs) are progressive disorders that include Ataxias, Alzheimer's, disease, Amyotrophic lateral sclerosis, Huntington's disease, Parkinson's disease, Multiple system atrophy and Progressive supranuclear palsy that are characterised by alteration of the structure and function of the peripheral and central nervous system resulting in nerve cell death [1]. The global transition in our demographics, which is characterised by an increasing elderly population [2-4] and compounded by the emerging long-term side effects of the COVID-19 pandemic has resulted in a global increase in the prevalence of NDDs [2, 5]. This increased prevalence coupled with the overwhelming burden of NDDs on individuals, families, and communities [3] is necessitating an increased urgency of developing novel therapies that would better manage

the diseases and also address the social and public health burden of NDDs. Generally, the aetiology of NDDs have been attributed to a number of factors including environment, genetics and lifestyle changes [4, 6]. However, individual disorders like Alzheimer's disease also have additional causes including neuroinflammation, oxidative stress, neurotransmitter dysfunction and energy dysmetabolism [1. 5, 7, 8].

Alzheimer's disease (AD), which is the most prevalent form of age-related dementias is characterised by progressive loss of spatial awareness, memory loss and personality changes [7]. The increasing social and economic burden associated with the development of AD has continued to make it a focus of extensive preclinical and clinical research. Recent advances in our understanding of the pathophysiological mechanisms of AD has been aided by the use of animal models which also allow the assessment of novel therapies These approaches which include the use of transgenic animal models, endogenous substance model, chemical models and heavy metal models of AD [9].

Heavy metal-induced models of AD have been developed based on the premise that exposure to heavy metals including cadmium, lead, manganese, fluoride and aluminium; a number of which are ubiquitous within environment or earth's crust but have been associated with increased risk of AD in the general population [9, 10].

Aluminium (Al) is a heavy metal which is widely distributed within the earth's crust and upon exposure readily crosses the blood brain barrier [9]. However, despite being ubiquitous in the environment, for aeons exposure to it has only been associated with deleterious effects in the body particularly in the brain [11]. Aluminium exposure has been associated with such as increased inflammation, oxidative stress amyloid β deposition, calcium dysmetabolism and increased formation of neurofibrillary tangles in the brain [11]; which are pathological hallmarks of AD. that are associated with the progression of AD [12]. Exposure to Al has also been linked with cholinergic and synaptic dysfunction that is now associated with memory impairment [9]. Several studies have demonstrated that prolonged exposure to Al is associated with accumulation of the heavy metal within the brain particularly the hippocampus and the induction of neuroinflammatory cytokines and oxidative stress [13-18] affirming its validity as a model for studying AD; however, inability to readily reproduce deposition of neurofibrillary tangles and amyloid-β plaques in this model could be a limitation to its use [19]. The use of AD disease models has enabled further understanding of the pathophysiological mechanism of AD. In addition to the previously known pathophysiological mechanisms, there are reports that have also linked the development of AD to deficits in the intracellular levels of pyridoxal 5'-phosphate [20, 21].

Pyridoxal 5'-phosphate (PLP) is the biochemically active form of vitamin B6 and is often required for biochemical reactions and synthesis of several neurotransmitters including dopamine, epinephrine, norepinephrine, serotonin, and gamma (γ) aminobutyric acid (GABA) [22-25]. In developing economies, the reduction in the levels of PLP is often due to dietary insufficiency, especially in infants, children, and women [26-28]. While several studies have demonstrated the ability of PLP to reverse neurodegenerative changes, oxidative stress and neuroinflammatory changes [29-31], there is however a dearth of scientific information on its effect in aluminium chloride model of AD, hence this study. This study examined the effects of pyridoxal phosphate supplementation on aluminium chloride induced neurobehavioural, and histomorphological changes in the cerebral cortex of rats.

2. **Materials and Methodology**

2.1 Chemicals and Drugs

Pyridoxal phosphate (Nature's field, USA), Aluminium chloride (Sigma-Aldrich Corporation, USA), normal saline, assay kits for lipid peroxidation (Malondialdehyde (MDA), interleukin-6 (IL-6), interleukin-1β (IL-1β), Tumour necrosis factor alpha (TNF-α), and total antioxidant capacity (TAC) (Biovision Inc., Milpitas, CA, USA).

2.2 Animals

Forty male Wistar rats were obtained from the Empire Breeders, Osogbo, Osun State and housed in wooden cages at room temperature (25°C±2.5°C) with 12 hours of light. Animals were acclimatised to the experimental conditions for two weeks.

2.3 Experimental Methods

Forty male Wistar rats weighing 130-150g each were distributed into four groups (n=10). Group A received distilled water (10 ml/kg), group B received $AlCl_3$ (50 mg/kg), group C and D received $AlCl_3$ and PLP at 100 mg/kg and 200 mg/kg respectively. Pyridoxal phosphate was administered on days 1-15 while $AlCl_3$ was administered on days 16-30. All agents were administered orally throughout the experiment. On the 31st day, animals were subjected to neurobehavioural paradigms including the open field box and Y-maze. On the 32nd day, animals were sacrificed by cervical dislocation and blood samples were taken via cardiac puncture for the assay of Total Antioxidant Capacity (TAC), Malondialdehyde (MDA), Tumour Necrosis Factor Alpha (TNF-α), Interleukin-1β (IL-1β) and Interleukin-6 (IL-6). The cerebral cortex was excised and processed for histological analysis using haematoxylin and eosin, and cresyl fast violet stains.

2.4 Behavioural Testing

At the end of the experimental period, animals were exposed to behavioural paradigms (open field arena and Y-maze) as previously described [32-35]. Behavioural tests were carried out daily between 7.00 am and 1.00 pm.

2.4.1 Open field Novelty induced Behaviours

Animals were exposed to the open field arena for ten minutes during which the following behaviours were observed and scored; horizontal locomotion, rearing and grooming. The open-field apparatus used was a square box with dimensions of 75 x 75 x 30 cm, featuring a hard floor. The interior of the box was divided into 16 equal squares by permanent red markings. Each rat was placed in the centre of the open field, and its behaviour was recorded as described [36, 37]. The recorded parameters within the 10-minute observation period included: total horizontal locomotion (number of floor units entered with all paws), rearing frequency (number of times the rat stood on its hind legs), and frequency of grooming (number of body-cleaning actions, licking of the body and pubis, and face-washing) which are indicative of stereotypic behaviour.

2.4.2 Memory (Y maze)

Spontaneous alternation is a measure of spatial working-memory. The Y-maze was used to measure short term memory, general locomotor activity and stereotypic behaviours. For each animal, the Y-maze testing was carried out for 5 minutes. The Y-maze consisted of three equally spaced arms, each with dimensions of 41cm in length and 15cm in height. Each arm's floor measured 5 centimeters in width. Observing spontaneous alternation behaviour which is a reflection of rodents' inclination to switch between non-reinforced options was used to gauge spatial working memory. After being put in one of the Y-maze's arm compartments, each rat was free to roam about until its tail fully entered another arm. The arms were labelled A, B, or C, and the arm entry sequence was manually recorded. Entry into each of the three arms in succession was referred to as an alternation. The total number of arms entered minus two was used to determine the number of maximal spontaneous alternations. Next, (actual alternations/maximum alternations) x 100 was used to get the percentage alternation. Between sessions, the equipment was cleaned with 5% alcohol following each test [38].

2.5 Biochemical Test

2.5.1. Interleukin (IL) -1β and Interleukin-6

Interleukin (IL)-1β and Interleukin-6 levels were measured using enzyme-linked immunosorbent assay methods with commercially available kits procured from Biovision Inc., Milpitas, CA, USA.

2.5.2. Antioxidant Status

Total antioxidant capacity was measured using the Trolox Equivalent Antioxidant Capacity Assay that is based on the ability of antioxidants within a sample to react with oxidised products as previously described [15, 16].

2.5.3. Lipid Peroxidation

Lipid peroxidation levels were assessed by determining the malondialdehyde (MDA) content, which assays the levels of thiobarbituric acid reactive substance in samples. Thiobarbituric acid reactive substance combine with free malondialdehyde present in samples to form a coloured complex, the concentration of which is expressed as μmol [15, 16, 37].

2.6 Tissue Histology

Rat brains were dissected, sectioned and fixed in neutral buffered formol calcium. The prefrontal cortex was then processed for paraffin-embedding, cut at 5 μm and stained with haematoxylin/eosin and cresyl fast violet for general histological study.

2.7 Photomicrography:

Histological slides of the prefrontal cortex were examined under an Olympus binocular light microscope. Images were captured using a Canon PowerShot 2500 digital camera. Histopathological alterations were evaluated by a pathologist who was unaware of the group assignments.

2.8 Statistical Analysis

Data was analysed using Chris Rorden's ANOVA for Windows (version 0.98). Analysis of data was by One-way analysis of variance (ANOVA) and a post-hoc test (Tukey HSD) used comparisons. All results were expressed as mean ± S.E.M. and $p < 0.05$ was taken

3. Results

3.1 Effects of pyridoxal phosphate on relative body and brain weights.

Figure 1 shows the effect of pyridoxal phosphate on changes in bodyweight. There was a significant decrease in body weight in the groups administered aluminium chloride ($AlCl_3$), $AlCl_3$ with pyridoxal phosphate (PLP) at 200 mg/kg and an increase with $AlCl_3$/PLP at 100 mg/kg compared to control. Compared to $AlCl_3$ control, relative body weight increased with $AlCl_3$/PLP at 100 and 200 mg/kg respectively.

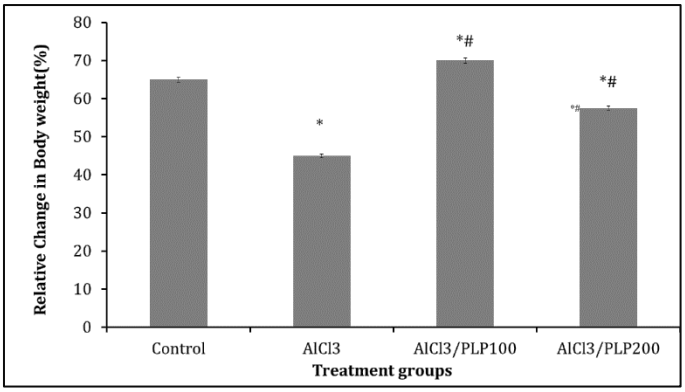

Figure 1: Shows the effect of pyridoxal phosphate on relative change in body weight. Each bar represents Mean ± S.E.M *p< 0.05 vs. control, #p<0.05 significant difference from $AlCl_3$, number of rats per treatment group =10. $AlCl_3$: Aluminium chloride, PLP: Pyridoxal Phosphate.

3.2 Effects of pyridoxal phosphate on relative food intake

Figure 2 shows the effect of pyridoxal phosphate on relative change in food intake. There was a significant decrease in food intake with $AlCl_3$, $AlCl_3$PLP at 100 and 200 mg/kg compared to control. Compared to $AlCl_3$ control, relative food intake increased with $AlCl_3$/PLP at 100 and 200 mg/kg respectively.

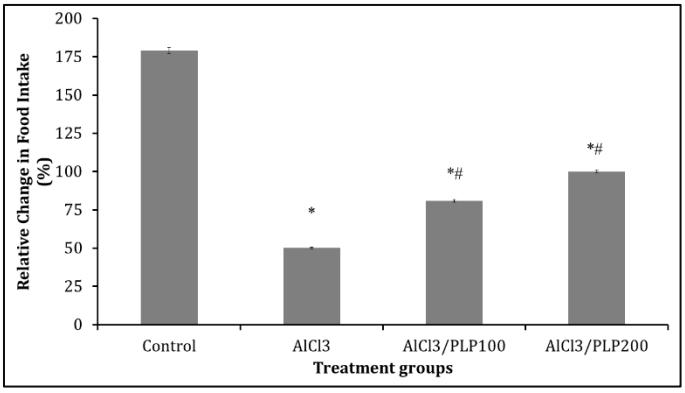

Figure 2: Shows the effect of pyridoxal phosphate on relative change in food intake. Each bar represents Mean ± S.E.M *p< 0.05 vs. control, #p<0.05 significant difference from $AlCl_3$, number of rats per treatment group =10. $AlCl_3$: Aluminium chloride, PLP: Pyridoxal Phosphate

3.3 Effect of pyridoxal phosphate on relative brain weight

Figure 3 shows the effect of pyridoxal phosphate on relative brain weight. There was no significant difference in relative brain weight in any of the groups compared to control or $AlCl_3$ control.

3.5 Effect of Pyridoxal Phosphate on rearing

Figure 5 shows the effect of pyridoxal phosphate on rearing activity in the open field arena. There was a significant decrease in rearing activity with $AlCl_3$, $AlCl_3$PLP at 100 and 200 mg/kg compared to control. Compared to $AlCl_3$ control, rearing activity increased with $AlCl_3$/PLP at 100 and 200 mg/kg respectively.

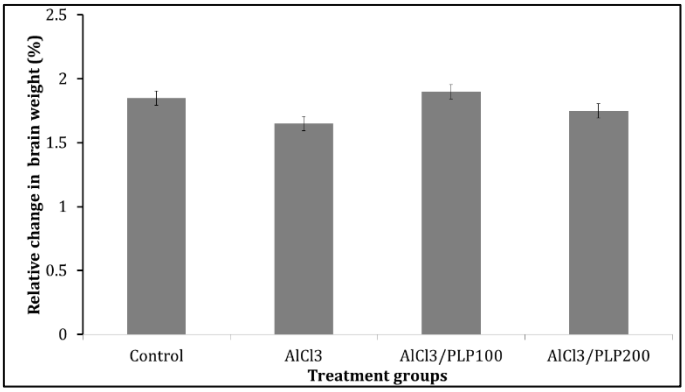

Figure 3: Shows the effect of pyridoxal phosphate on relative brain weight. Each bar represents Mean ± S.E.M, number of rats per treatment group =10. $AlCl_3$: Aluminium chloride, PLP: Pyridoxal Phosphate.

3.4 Effect of Pyridoxal phosphate on locomotor activity

Figure 4 shows the effect of pyridoxal phosphate on horizontal locomotion in the open field arena. There was a significant decrease in locomotor activity with $AlCl_3$, $AlCl_3$PLP at 100 and 200 mg/kg compared to control. Compared to $AlCl_3$ control, locomotor activity increased with $AlCl_3$/PLP at 100 and 200 mg/kg respectively.

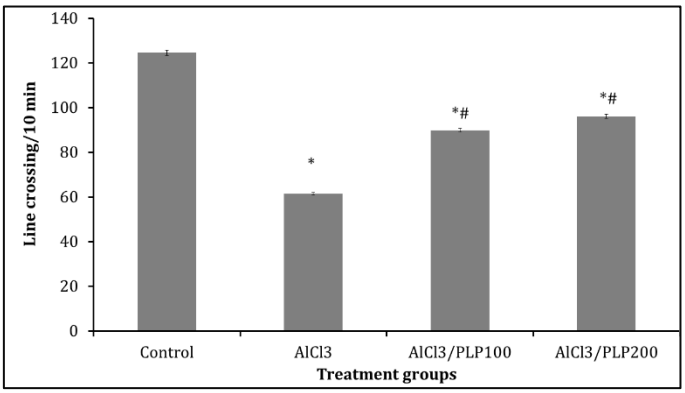

Figure 4: Shows the effect of pyridoxal phosphate on horizontal locomotion in the open field arena. Each bar represents Mean ± S.E.M *p< 0.05 vs. control, #p<0.05 significant difference from $AlCl_3$, number of rats per treatment group =10. $AlCl_3$: Aluminium chloride, PLP: Pyridoxal Phosphate

3.6 Effect of Pyridoxal Phosphate on self-grooming

Figure 6 shows the effect of pyridoxal phosphate on self-grooming behaviour in the open field arena. There was a significant decrease self-grooming behaviours with $AlCl_3$, and $AlCl_3$PLP at 200 mg/kg and a significant increase with $AlCl_3$PLP at 100 mg/kg compared to control. Compared to $AlCl_3$ control, self-grooming behaviour increased with $AlCl_3$/PLP at 100 and decreased with $AlCl_3$PLP at 200 mg/kg.

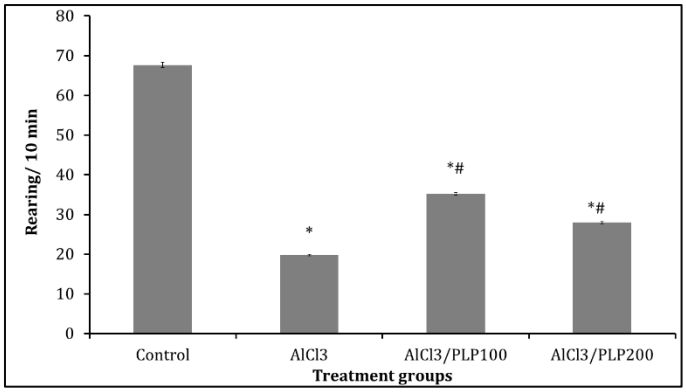

Figure 5: Shows the effect of pyridoxal phosphate on rearing activity in the open field arena. Each bar represents Mean ± S.E.M *p< 0.05 vs. control, #p<0.05 significant difference from $AlCl_3$, number of rats per treatment group =10. $AlCl_3$: Aluminium chloride, PLP: Pyridoxal Phosphate.

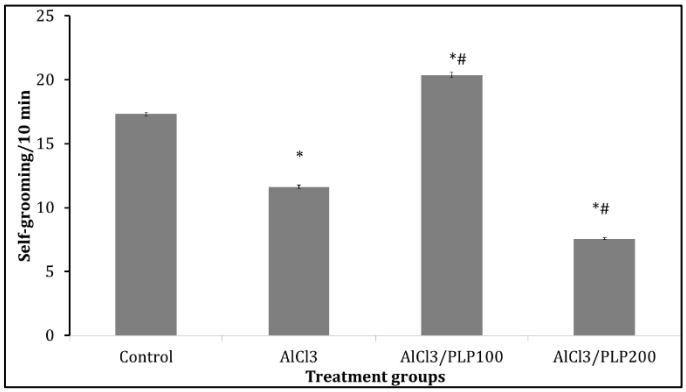

Figure 6: Shows the effect of pyridoxal phosphate on self-grooming behaviour in the open field arena. Each bar represents Mean ± S.E.M *p< 0.05 vs. control, #p<0.05 significant difference from $AlCl_3$, number of rats per treatment group =10. $AlCl_3$: Aluminium chloride, PLP: Pyridoxal Phosphate

3.7 Effect of pyridoxal phosphate on working memory

Figure 7 shows the effect of pyridoxal phosphate on spatial working memory in the Y-maze measured as % alternation. There was a significant decrease working memory with $AlCl_3$,, $AlCl_3$PLP at 100 and 200 mg/kg compared to control. Compared to $AlCl_3$ control, working memory increased with $AlCl_3$/PLP at 100 and 200 mg/kg.

3.8 Effect of pyridoxal phosphate on oxidative stress parameters and inflammatory cytokines

Table 1 shows the effects of pyridoxal phosphate shows the effect of pyridoxal phosphate on oxidative stress parameters and inflammatory cytokines. Antioxidant status measured as Total antioxidant capacity (TAC) decreased significantly with $AlCl_3$ and $AlCl_3$PLP at 100 mg/kg and increased significantly with $AlCl_3$PLP at 200 mg/kg compared to control. Compared to $AlCl_3$ control, TAC levels increased with $AlCl_3$/PLP at 100 and 200 mg/kg. respectively

Oxidative stress measured as malondialdehyde (MDA) increased significantly with $AlCl_3$ and $AlCl_3$PLP at 100 mg/kg and decreased significantly with $AlCl_3$PLP at 200 mg/kg compared to control. Compared to $AlCl_3$ control, MDA levels decreased with $AlCl_3$/PLP at 100 and 200 mg/kg. respectively.

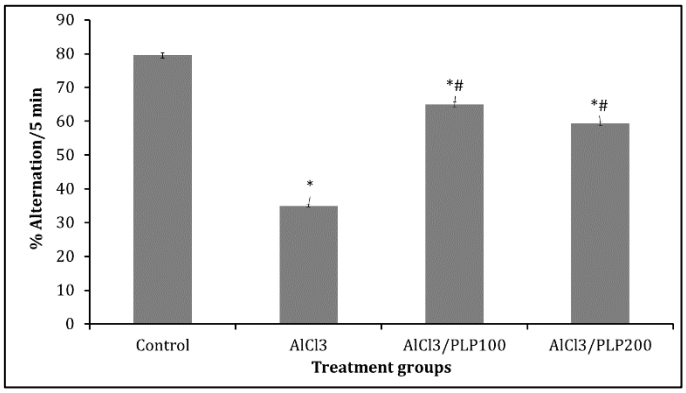

Figure 7: Shows the effect of pyridoxal phosphate on spatial working memory in the Y maze. Each bar represents Mean ± S.E.M *p< 0.05 vs. control, #p<0.05 significant difference from $AlCl_3$, number of rats per treatment group =10. $AlCl_3$: Aluminium chloride, PLP: Pyridoxal Phosphate.

Inflammatory cytokines measured were interleukin (IL)-1β, IL-6 and Tumour necrosis factor (TNF)-α. interleukin (IL)-1β, IL-6 and TNF -α levels increased significantly with $AlCl_3$ and $AlCl_3$PLP at 100 and 200 mg/kg compared to control. Compared to $AlCl_3$ control, levels of all three inflammatory cytokines decreased with $AlCl_3$/PLP at 100 and 200 mg/kg. respectively

Table 1: Effect of pyridoxal phosphate on oxidative stress and proinflammatory parameters

Groups	TAC (mM)	MDA (µM)	IL-6 (pg/ml)	IL-1β (pg/ml)	TNF-α(pg/ml)
Control	18.92 ±0.10	5.17 ±0.10	2.68 ±1.20	6.94 ±1.00	19.04 ±0.34
$AlCl_3$	9.81 ±0.12*	14.0 ±0.12*	15.23 ±1.80*	17.99 ±2.20*	56.59 ±0.65*
$AlCl_3$/PLP 100	15.80 ±0.12*#	9.72 ±0.14*#	10.94 ±2.15*#	17.97 ±2.20*	30.56 ±0.15*#
$AlCl_3$/PLP 200	24.65 ±0.15*#	3.25 ±0.30*#	7.64 ±2.60*#	10.47 ±2.20*#	26.5 5±0.30*#

Mean ± S.E.M, *p<0.05 significant difference from control. #p<0.05 significant difference from $AlCl_3$. Number of rats per treatment group =10. $AlCl_3$: Aluminium chloride, PLP: Pyridoxal phosphate, TNF-α: Tumor necrosis factor α. Il: Interleukin.

3.9 Effect of pyridoxal phosphate on the cerebral cortex histomorphology

Figure 8 (A-D) and 9 (A-D) represent the photomicrographs of haematoxylin and eosin (H&E) and cresyl fast violet (CFV) stained sections of the rat cerebral cortex respectively. Examination of the H&E-stained slides of rats in the control group revealed characteristic features of the inner pyramidal layer of the cortex. The CFV stain micrographs also revealed well preserved multipolar shaped pyramidal cells within the neuropil. Granular neurons with large open-faced nuclei, scanty cytoplasm and small sized neuroglial cells were also observed.

In the group administered $AlCl_3$, numerous degenerating pyramidal and granule cells and giant cells with double nuclei were observed features which are in keeping with neuronal degeneration and neuronal injury. Also observed were pale-staining neurons with shrunken nuclei. which were interspersed between a few normal pyramidal and granule cells. In the groups administered $AlCl_3$/PLP 100 mg/kg (8C & 9 C) and $AlCl_3$/PLP 200 mg/kg (8 D and 9 D), reversal of neuronal injury and preservation of Nissl bodies were observed compared with the $AlCl_3$ group.

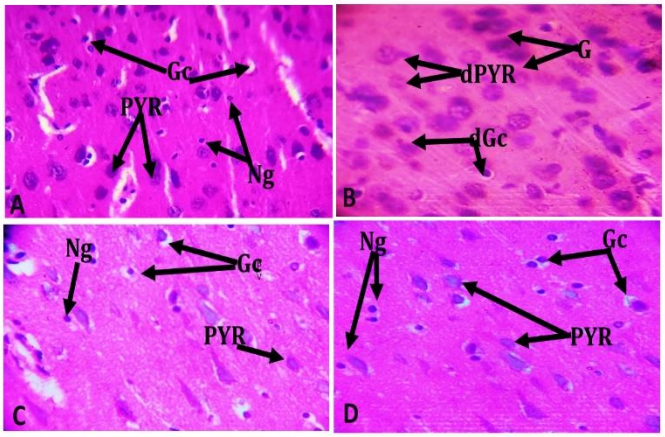

Figure 8: Representative photomicrograph of haematoxylin and Eosin-stained sections of the rat cerebral cortex. Photomicrographs revealed distinct layers of the cerebral cortex with presence of numerous granule cells (Gc), pyramidal cells (PYR), and neuroglial cells (NG). Also observed are degenerating pyramidal cell (dPYR), degenerating granule cell (dGc) and Giant cell G. Magnification: x400.

4. Discussion

This study investigated the possible protective effects of pyridoxal phosphate on aluminium chloride-induced neurobehavioural, biochemical and histomorphological changes in rats. The results revealed that pyridoxal phosphate protected against neurotoxic effects associated with aluminium chloride ingestion supporting other literature that had reported similar findings following aluminium chloride ingestion in rodents [15, 16, 39]- Significant changes were observed in food consumption, relative body weight, neurobehavioural parameters, antioxidant status, oxidative stress and proinflammatory markers. In this study, animal exposure to $AlCl_3$ revealed a significant reduction in feed intake, body weight, antioxidant capacity and behavioural parameters. There was also an increase in the levels of MDA, IL-1β, IL-6 and TNF-α in the $AlCl_3$ group compared to the control. Photomicrographs of the $AlCl_3$ group further revealed neuronal degeneration in the histopathological architecture of the brain.

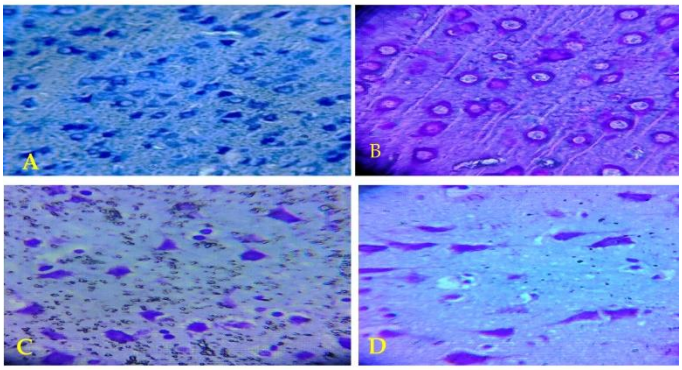

Figure 9: Photomicrograph of cresyl violet stained sections of the rat cerebral cortex section by cresyl fast violet X100. The photomicrographs show the neuronal cell scattered within the Nissl substance in groups A (Control), C ($AlCl_3$ and pyridoxal Phosphate at 100 mg/kg) and D ($AlCl_3$ and pyridoxal Phosphate at 200 mg/kg) and neurodegenerative cells in group B (Aluminium-chloride control). Magnification x 400.

The administration of PLP however, protected against the development of the deleterious effects of $AlCl_3$ across different parameters including feed intake, body weight, neurobehaviour, oxidative stress marker sand proinflammatory cytokines. The photomicrographs of the treated group further revealed a reversal of the damage to the pyramidal and granular neurons as observed in the $AlCl_3$ group. The reduction seen in feed intake and body weight following the administration of $AlCl_3$ corroborates reports of previous studies that had reported significant reduction in feed intake and body weight following exposure of animals to $AlCl_3$ [15, 16]. Moreover, the significant increase in weight of animals treated with $AlCl_3$/PLP100 mg/kg and $AlCl_3$/PLP200mg/kg, corroborates reports of the research findings by Olofinnade *et al* [29] that examined the effects of pyridoxal phosphate in chlorpromazine-induced parkinsonism rodent model. They reported that PLP administration at 100 mg/kg was associated with weight gain [29] similar to our observations in this study.

Furthermore, blood assays revealed a significant increase in the total antioxidant capacity (TAC) levels with pyridoxal phosphate treated group compared with $AlCl_3$ group while MDA, IL-6 and TNF-α levels decreased significantly with the treated group compared with $AlCl_3$ group. There was also a significant decrease in IL-1β level in the group treated with $AlCl_3$/PLP 200 compared with groups $AlCl_3$ and $AlCl_3$/PLP100. Exposure to $AlCl_3$ have been reported by previous studies to

significantly elevate brain concentrations of some key pro-inflammatory cytokines like TNF-α, IL-1β and IL-6 [15,16]. The upregulation of these cytokines stimulates leukocyte production which will further intensify the inflammatory response leading to more production of additional pro-inflammatory mediators to the site of call. According to Abulfadl *et al.* [40] accumulation of $AlCl_3$ triggers the release of cytochrome C from mitochondria, leading to an increased free radical production and elevated pro-inflammatory cytokine levels [41]. In this study PLP's ability to increase antioxidant status was observed which was evidenced by a reduction in MDA and an increase in TAC levels in groups that received PLP, corroborating the evidence from previous studies that demonstrated PLP's significant antioxidant effects [42-45]. These neuroprotective effects of PLP have also been linked to its ability to inhibit the formation of malondialdehyde and scavenge reactive oxygen species [44, 45]. In addition, PLP's ability to reduce lipid peroxidation, increase antioxidant levels, and decrease inflammation even at the molecular level are possible mechanisms through which it reversed the damaging effects of $AlCl_3$ [14].

Exploration of the neurobehavioural paradigms revealed a significant increase in the open field exploration activities in the treated group compared with $AlCl_3$ group. Previous studies had reported that animal exposed to $AlCl_3$ exhibited behaviours like impaired locomotion, self-grooming, and anxiety-like responses This is as a result of $AlCl_3$'s interference with cholinergic and dopaminergic systems that play vital roles in locomotion, memory and self-awareness. Dopaminergic and cholinergic dysfunction are key to motor and self-maintenance deficits [46]. The increase observed in the open field exploration activities in the PLP treatment groups compared to $AlCl_3$ group suggests that PLP has a modulatory effect on locomotion, rearing and grooming. According to Olofinnade *et al* [29], PLP modulated behaviours possibly through its positive effects on brain neurotransmitters activity or receptor binding. Besides, the improved performance observed in the exploration activities in the Y-maze apparatus of the $AlCl_3$/PLP groups compared to $AlCl_3$ group also supported the work done by Jeanclos *et al* [47] and Olofinnade *et al* [29], that reported an increase in the levels of pyridoxal phosphate was associated with increased cognition.

Lastly, aluminium chloride administration had been reported to be associated with morphological evidence of neuronal injury due to its neurotoxic effects [15, 16]. Histopathological analysis using H&E and cresyl fast violet stains further corroborated these findings. In this study, the photomicrographs of animals exposed to $AlCl_3$ revealed notable cortical degeneration, loss of neuronal structure and depletion of Nissl body substances. This indicates possible disruption in protein synthesis and neuronal function. However, animals treated with pyridoxal phosphate revealed a preserved brain architecture, nearly restoring normal pyramidal cell morphology and Nissl body presence.

Conclusion

In conclusion, this study demonstrated the possible protective effects of pyridoxal phosphate supplementation on aluminium chloride induced changes in behaviour, oxidative stress, proinflammation and cerebral cortex histomorphology in rats. However, more studies are required to determine its benefits in humans.

Funding.
None

Availability of data and materials

Data generated during and analysed during the course of this study are available from the corresponding author on request.

Declarations Ethics approval

Ethical approval for this study was granted by the Ethical Committee of the Faculty of Basic Medical Sciences.

Competing interests

All authors of this paper declare that there is no conflict of interest related to the content of this manuscript.

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