OPEN ACCESS

Saudi Journal of Medical and Pharmaceutical Sciences
Abbreviated Key Title: Saudi J Med Pharm Sci
ISSN 2413-4929 (Print) | ISSN 2413-4910 (Online)
Scholars Middle East Publishers, Dubai, United Arab Emirates
Journal homepage: https://saudijournals.com

Original Research Article **Physiology**

Cognito-Motor Modulatory Functions of Lutein on Diazepam-Induced Memory Impairment in Male Wistar Rats

Austin. A. Ajah[1]*, Ekoriko P[2]

[1]Department of Physiology, Faculty of Basic Medical Sciences, College of Health Sciences, University of Port Harcourt, P.M.B. 5323, Choba, Port Harcourt, Nigeria
[2]Department of Medical Biochemistry, Faculty of Basic Medical Sciences, College of Health Sciences, University of Port Harcourt, P.M.B. 5323, Choba, Port Harcourt, Nigeria

DOI: https://doi.org/10.36348/sjmps.2025.v11i11.019 | **Received:** 02.10.2025 | **Accepted:** 24.11.2025 | **Published:** 26.11.2025

***Corresponding author:** Austin. A. Ajah
Department of Physiology, Faculty of Basic Medical Sciences, College of Health Sciences, University of Port Harcourt, P.M.B. 5323, Choba, Port Harcourt, Nigeria

Abstract

Lutein as a carotenoid, provides a wide range of nutritional and health benefits due to their multifarious biological impacts on humans, which include antioxidative, immunomodulatory, and anti-inflammatory properties. This study was carried out to evaluate the Cognito motor modulatory function of lutein on Hippocampal cellular Architecture in Diazepam induced memory impairment in Wistar rats. Thirty (30) male rats were used for this study and the rats were acclimatized for a period of 14 days, and was then divided into six groups; 1 (Control), 2 (Diazepam 5mg/kg), 3 (Diazepam + Lutein 20mg/kg), 4 (Diazepam + Lutein 40mg/kg), 5 (Diazepam + Lutein 60mg/kg), and 6 (Diazepam + Donepezil - Standard drug) for a period of 21 days. The catalase level was seen to be statistically significant ($p<0.05$) when the group 2 (diazepam only treated) and group 3 were separately compared to that of the control group (group 1). In neurobehavioural test conducted, Group 2 in handgrip stability time test conducted showed a significant ($p<0.05$) decrease signifying a lack of peak force for handgripping when compared with group 1(control group), this was made significant by the increase in the number of incorrect hole entry by group 2 wistar rats in Barnes maze test in week 1 and 2 of the study period, Lutein showed a motor modulatory function in the brain through its antioxidizing and anti-proteolytic properties, with its dose dependence effect on group 3 Wistar rats, of which catalase concentration level seems to be lesser when compared to group 2.
In summary, lutein showed dose dependence effect in motor modulatory function of the brain through its antioxidizing and anti-proteolytic properties,

Keywords: Cognito-Motor, Lutein, Hippocampus, Diazepam, Memory Impairment.

INTRODUCTION

There are nearly about 10 million new cases every year pertaining to impaired memory, making it one of the major causes of disability and dependency among older people worldwide (WHO, 2025). The morbidity and mortality rates of memory impairment are both quickly growing in both developed and undeveloped countries (Amiri *et al.,* 2025). Over 50 million people are living with some sort of memory impairment worldwide (WHO, 2025). These numbers are projected to reach 82 million by the year 2030 and 152 million by 2050, with the majority of individuals coming from low- and middle-income countries (Guerchet *et al.,* 2020; WHO, 2021).

In Nigeria, about 5-8% of general population aged 60 and above is living with some sort of memory impairment (Yarube, 2021). Apart from dementia in its various forms such as Alzheimer's disease and vascular dementia, several other disease conditions may present with cognitive impairment, and many of these conditions have a high prevalence in Nigeria (Yarube, 2021). These conditions form the sources and ramifications of cognitive impairment. They include aging population (Ibrahim Abdu Wakawa *et al.,* 2025), diabetes mellitus (both type 1 and type 2), hypertension, stroke, Parkinson's disease, depression, pregnancy and HIV/AIDS; and possibly other unconfirmed conditions associated with cognitive impairment such as sickle cell disease, due to vascular dementia, head injury, nutritional deficiencies (Jamalnia *et al.,* 2020),

communal conflict and internal displacement (Yarube, 2021). When the contributions of these different conditions add up, the prevalence figure for cognitive impairment and dementia will be staggering (Yarube, 2021).

This high local burden of cognitive impairment is a source of concern and a call to finding a local solution to the problem through research and innovation. This is necessary for policy making and healthcare delivery planning regarding cognitive impairment and dementia in the country.

This research will determine the effect of administration of lutein on Cognito-motor modulatory functions. The long-term administration of diazepam on Wistar rats and the effect of administration of different doses of lutein on neurobehavioral test for memory impaired Wistar rats will be evaluated. Wistar rats as mammals have similar central nervous system as humans. The influence of lutein on the Wistar rats can have almost the same effect on human with specific consideration of dosage and weight.

MATERIALS AND METHODS

Research Design

This animal-based study was conducted at the Animal House, Department of Human Physiology, Faculty of Basic Medical Sciences, University of Port Harcourt. The experiment lasted five weeks (35 days), including a 2-week acclimatization period and 3-week drug administration period.

Thirty (30) male Wistar rats (111-148g) were randomly divided into six groups (n = 5 per group). Group 1 served as the control, receiving distilled water and normal feed. Groups 2-6 were test groups, receiving various combinations of diazepam, lutein, dimethyl sulfoxide ($DMSO_4$), and donepezil.

Drug administration was carried out daily for 21 consecutive days (08:00-09:00 a.m). Diazepam was administered intraperitoneally, followed one hour later by oral administration of lutein suspensions or donepezil, depending on the group. Neurobehavioral tests were conducted 30 minutes after lutein administration.

Ethical Approval

Ethical approval was obtained from the faculty of basic medical science, Abuja campus, University of Port Harcourt. Rat handling and treatment conform to the guideline of the National Research Council (2011) for care and use of laboratory animals.

Drug Preparation

Diazepam, lutein, donepezil, and $DMSO_4$ were obtained from the National Institute for Pharmaceutical Research and Development (NIPRD), Rivers State, Nigeria, and confirmed by the Department of Pharmacy, University of Port Harcourt. Lutein suspensions were prepared using the freezing and overnight dilution method.

Drug Administration

Drugs were prepared by suspending the appropriate weights of lutein and $DMSO_4$ in distilled water. Administration was carried out orally using a 2ml syringe with cannula. Diazepam was given intraperitoneally. All doses were calculated based on the average weekly body weight of each group.

Table 1: Experimental Grouping and Drug Administration

Group	Treatment	Average Weight Range (g) (Week 1 -3)	Dose Administered	Route of Administration
1	Control (normal feed + distilled water)	140-160		Oral (feed/water)
2	Diazepam (5 mg/kg)	140-160	0.1 mg/kg	Intraperitoneal (IP)
3	Diazepam (5 mg/kg) + Lutein (20 mg/kg) + $DMSO_4$ (2 mL in 8 mL water)	140-160	Diazepam 0.1 mg/kg + Lutein 0.9 mg/kg	IP + Oral
4	Diazepam (5 mg/kg) + Lutein (40 mg/kg) + $DMSO_4$ (2 mL in 8 mL water)	140-160	Diazepam 0.1 mg/kg + Lutein 0.8-1.0 mg/kg	IP + Oral
5	Diazepam (5 mg/kg) + Lutein (60 mg/kg) + $DMSO_4$ (2 mL in 8 mL water)	140-160	Diazepam 0.1 mg/kg + Lutein 0.8-0.9 mg/kg	IP + Oral
6	Diazepam (5 mg/kg) + Donepezil (10 mg/kg)	140-160	Diazepam 0.1 mg/kg + Donepezil 1.2 mg/kg	IP + Oral

Neurobehavioral Tests

A number of behavioral tests was conducted to assess motor coordination, spatial memory, anxiety, and exploratory behavior.

- **Hand Grip Test** - Measured forelimb grip strength using a grip strength meter and wire mesh/wire hang apparatus.
- **Rotarod Test** - Measured motor coordination and balance. Rats were trained for 7 days prior to testing; test trials lasted 120 s at 15 rpm.
- **Y-Maze Test** - Evaluated acquisition of spatial memory using a three-arm maze. Parameters recorded: spontaneous alternation and inflexion ratio.
- **Barnes Maze Test** - Evaluated locomotor activity, exploration, and anxiety. Parameters: total distance traveled, time spent in center vs. periphery, rearing, and grooming.

Biochemical Analysis

Brain tissue homogenates were analyzed for Catalase levels by using Aebi Method, which involves monitoring the decrease in hydrogen peroxide ($H2O_2$) absorbance at 240 nm in a spectrophotometer (Khovarnagh & Seyedalipour, 2021).

Statistical Analysis

Statistical analysis was conducted using GraphPad 8 software and the results were expressed as Mean ± SEM. Group comparisons were made using one-way ANOVA, followed by least significant difference (LSD) post hoc test. Statistical significance was accepted at $p < 0.05$.

RESULTS

Table represent Mean ± SEM, n=5; a Significant at p<0.05 compared to Group 1; b Significant at p<0.05 when compared to group 2; c Significant at p<0.05 when compared to group 3. group 4, group 5, and group 6.

Table 2: Effect of Lutein Administration on Catalase Concentration Level in Female Wistar Rats

Variables	Groups					
	Control	Diazepam Only	Diazepam + Lutein (20 mg/kg)	Diazepam + Lutein (40 mg/kg)	Diazepam + Lutein (60 mg/kg)	Diazepam + Donepenzil (10mg/kg)
Catalase (CAT)	0.29 ± 0.05	1.51 ± 0.12 ****	0.77 ± 0.06 **	0.62 ± 0.03	0.51 ± 0.09	0.33 ± 0.09

Table 3: Effect of Lutein Administration on Neurobehavioural Test in Female Wistar Rats

Variables	Groups					
	Control	Diazepam Only	Diazepam + Lutein (20mg/kg)	Diazepam + Lutein (40mg/kg)	Diazepam + Lutein (60mg/kg)	Diazepam + Donepenzil (10mg/kg)
Rotarod Stability Time (s)	6.13 ± 0.89	1.47 ± 0.25 ***	3.47 ± 0.81	3.33 ± 0.37	4.24 ± 0.63	3.40 ± 0.92
Handgrip Stability Time (s)	3.67 ± 0.30	1.73 ± 0.12**	3.65 ± 0.34	4.12 ± 0.24	3.53 ± 0.23	3.72 ± 0.42
Y Maze Inflexion Ratio (s)	0.76 ± 0.13	1.60 ± 0.13****	1.11 ± 0.10	0.41 ± 0.11	0.39 ± 0.05	0.56 ± 0.04
Y Maze % Spontaneous Alteration	32.91 ± 3.26	10.22 ± 0.22****	14.20 ± 1.50****	34.50 ± 0.73	36.89 ± 1.12	32.11 ± 2.38
Barnes maze - Time spent in locating correct hole. Week 1	14.80 ± 1.39	27.00 ± 2.72**	17.60 ± 2.16	12.20 ± 1.20	15.00 ± 2.10	13.80 ± 3.03
Barnes maze - Time spent in locating correct hole. Week 2	10.40 ± 1.63	43.40 ± 7.51****	4.80 ± 1.53	7.80 ± 0.97	19.00 ± 2.82	8.40 ± 3.79
Barnes maze - Time spent in locating correct hole. Week 3	12.80 ± 3.81	42.20 ± 3.11****	30.20 ± 4.07*	6.20 ± 1.32	10.00 ± 4.11	12.60 ± 4.57
Barnes maze - Time spent in locating incorrect hole. Week 1	2.20 ± 0.37	5.20 ± 0.37**	3.40 ± 0.51	2.00 ± 0.45	2.60 ± 0.51	1.80 ± 0.49
Barnes maze - Time spent in locating incorrect hole. Week 2	0.60 ± 0.40	2.80 ± 0.37**	0.60 ± 0.40	1.00 ± 0.32	0.80 ± 0.37	0.80 ± 0.37
Barnes maze - Time spent in locating incorrect hole. Week 3	3.40 ± 1.16	2.80 ± 0.86	3.60 ± 0.81	1.80 ± 1.11	2.40 ± 0.93	3.88 ± 0.49

DISCUSSION

Antioxidant Enzyme Activity - Catalase Concentration Level

Table 2 indicates that lutein has significantly higher catalase (CAT) concentration in the treated groups than diazepam alone, which shows an increase in antioxidant defense. This correlates with prior results that lutein is a powerful antioxidant, which prevents oxidative stress in neural tissue by scavenging free radicals and increasing endogenous enzymes (Johnson, 2012; Li *et al.*, 2020). In comparable investigations, it has been pointed out that lutein supplementation enhanced biomarkers of oxidative stress in rodents with neurotoxicity models (Pap *et al.*, 2022).

Neurobehavioral Tests

1. **Handgrip and Rotarod Performance:** Lutein improves grip strength and neuromuscular

coordination when administered. As seen in table 3, there was a significant increase in handgrip stability time, and in in rotarod stability time, it was depicted that there was an improvement in rotarod performance. These findings are in line with others indicating that carotenoids, such as lutein, preserve the neuromuscular activity and suppress the motor impairments caused by neurotoxicants (Jayakanthan *et al.*, 2024).

2. **Y-Maze Performance:** In the Y-maze tests (Y maze inflexion ratio and Y maze % spontaneous alteration), lutein increased spatial working memory. Rats treated with lutein showed higher inflexion ratios and significantly better percentages of spontaneous alternation as compared to those rats treated with diazepam only. The current results agree with the previous rodent research that lutein supplementation enhances hippocampus-dependent memory formation (Nejad *et al.*, 2024).
3. **Barnes Maze Performance:** The Barnes maze test conducted in three weeks also supported the findings of lutein as a memory-enhancing substance. The latency of locating the correct hole was decreased in the lutein-treated groups and the number of incorrect entries was less. Such enhancements with time indicate that lutein can help in learning and long-term memory recall. Similar findings were also presented by Nazari *et al.*, (2022) who observed that lutein enhanced visual memory and cognitive flexibility in animal studies. Conversely, some studies show that the cognitive effects of lutein can be dosage and/or antioxidant dependent (Nejad *et al.*, 2024).

CONCLUSION

In summary, these results are indicative of the neuroprotective effects of lutein. It also boosts antioxidant defenses, enhances motor coordination and assists memory performance in cognitive impairment by diazepam. The observed effects, especially in the dose of 60mg/kg of lutein and the standard drug group; Diazepam (5 mg/kg) ± Donepezil (10 mg/kg), indicate that lutein may be used as an adjunct therapy in the treatment of neurodegenerative diseases like Alzheimer.

REFERENCES

- Amiri, F., Safiri, S., Shamekh, A., Ebrahimi, A., Sullman, M. J. M., & Kolahi, A.-A. (2025). Prevalence, deaths and disability-adjusted life years due to Alzheimer's disease and other dementias in Middle East and North Africa, 1990–2021. *Scientific Reports*, *15*(1). https://doi.org/10.1038/s41598-025-89899-w
- Guerchet, M., Prince, M., & Prina, M. (2020). Numbers of People with Dementia worldwide: an Update to the Estimates in the World Alzheimer Report 2015. *Www.alzint.org*. https://www.alzint.org/resource/numbers-of-people-with-dementia-worldwide/
- Ibrahim Abdu Wakawa, Umar Baba Musami, Suleiman Hamidu Kwairanga, Placidus Nwankuba Ogualili, Mahmood, M. Y., Muhammad Abba Fugu, Gimba, M. M., Muktar Mohammed Allamin, Abbas, Z. U., Muhammad Kawu Sunkani, Zainab Bukar Yaganami, Kadau, F. M., Sani, N. M., Danmallam, P., Luka Nanjul, Larema Babazau, Muhammad, Z., Goni, B. W., Babagana Kundi Machina, & Karch, C. M. (2025). Dementia in a resource-constrained sub-Saharan African setting: A comprehensive retrospective analysis of prevalence, risk factors, and management at the only neuropsychiatric facility in Northeastern Nigeria. *Alzheimer S & Dementia*, *21*(3). https://doi.org/10.1002/alz.14538
- Jamalnia, S., Javanmardifard, S., Akbari, H., Sadeghi, E., & Bijani, M. (2020). Association Between Cognitive Impairment and Blood Pressure Among Patients with Type II Diabetes Mellitus in Southern Iran. *Diabetes, Metabolic Syndrome and Obesity: Targets and Therapy*, *Volume 13*, 289–296. https://doi.org/10.2147/dmso.s238247
- Jayakanthan, M., Manochkumar, J., Efferth, T., & Ramamoorthy, S. (2024). Lutein, a versatile carotenoid: Insight on neuroprotective potential and recent advances. *Phytomedicine*, *135*, 156185. https://doi.org/10.1016/j.phymed.2024.156185
- Johnson, E. J. (2014). Role of lutein and zeaxanthin in visual and cognitive function throughout the lifespan. *Nutrition Reviews*, *72*(9), 605–612. https://doi.org/10.1111/nure.12133
- Khovarnagh, N., & Seyedalipour, B. (2021). Antioxidant, histopathological and biochemical outcomes of short-term exposure to acetamiprid in liver and brain of rat: The protective role of N-acetylcysteine and S-methylcysteine. *Saudi Pharmaceutical Journal*, *29*(3), 280–289. https://doi.org/10.1016/j.jsps.2021.02.004
- Li, L. H., Lee, J. C.-Y., Leung, H. H., Lam, W. C., Fu, Z., & Lo, A. C. Y. (2020). Lutein Supplementation for Eye Diseases. *Nutrients*, *12*(6), 1721. https://doi.org/10.3390/nu12061721
- National Research Council. (2011). *Guide for the Care and Use of Laboratory Animals, 8th edition. National Academies Press*. https://grants.nih.gov/grants/olaw/guide-for-the-care-and-use-of-laboratory-animals.pdf
- Nazari, L., Komaki, S., Salehi, I., Raoufi, S., Golipoor, Z., Kourosh-Arami, M., & Komaki, A. (2022). Investigation of the protective effects of lutein on memory and learning using behavioral methods in a male rat model of Alzheimer's disease. *Journal of Functional Foods*, *99*, 105319. https://doi.org/10.1016/j.jff.2022.105319
- Nejad, H. A., Nejad, A. Y., Akbari, S., Naseh, M., Mostafa, S., & Haghani, M. (2024). The low and high doses administration of lutein improves memory and synaptic plasticity impairment through different mechanisms in a rat model of vascular

dementia. *PLoS ONE*, *19*(5), e0302850–e0302850. https://doi.org/10.1371/journal.pone.0302850

- Pap, R., Pandur, E., Jánosa, G., Sipos, K., Nagy, T., Agócs, A., & Deli, J. (2022). Lutein Decreases Inflammation and Oxidative Stress and Prevents Iron Accumulation and Lipid Peroxidation at Glutamate-Induced Neurotoxicity. *Antioxidants*, *11*(11), 2269. https://doi.org/10.3390/antiox11112269
- WHO. (2021, September 2). *World failing to address dementia challenge*. World Health Organization. https://www.who.int/news/item/02-09-2021-world-failing-to-address-dementia-challenge
- WHO. (2025, March 31). *Dementia*. Who.int; World Health Organization: WHO. https://test-cms.who.int/westernpacific/newsroom/fact-sheets/detail/dementia
- Yarube, I. U. (2021). The burden of cognitive impairment in Nigeria: Perspectives and research prospects. *Journal of African Association of Physiological Sciences*, *9*(2), 69–73. http://www.jaaps.aapsnet.org.