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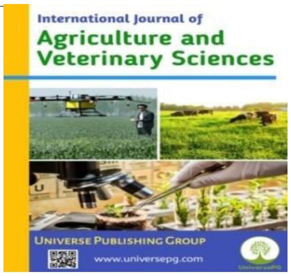

Effect of Dietary Tigernut on Acetylcholinesterase, Specific Acetylcholinester, and Total Protein Levels in Rabbit Brains of Different Genotypes

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ABSTRACT

This research investigated the impact of different levels of dietary *Cyperus esculentus* (tigernut) on acetylcholinesterase, specific acetylcholinesterase, and total protein concentrations in various brain regions of three rabbit genotypes. Male and female Dutch Belted, Hyla Max, and New Zealand White rabbits, totaling 320 at 6 weeks of age, were randomly assigned to diets containing 0, 10, 20, or 30g/kg of tigernut. Each treatment group was replicated five times with 15 rabbits per replicate. The rabbits were provided with ad-libitum feeding and clean water for 8 weeks or 56 days, after which six rabbits per replicate were euthanized. The brains were dissected into different regions including the olfactory lobe, pineal body, optic lobe, cerebellum, and medulla oblongata. The brain regions were homogenized to assess acetylcholinesterase and total protein concentrations, which were further used to evaluate specific acetylcholinesterase levels. The study found no significant differences in acetylcholinesterase activity in the olfactory lobe, pineal body, and cerebellum, except for the optic lobe and medulla. Additionally, dietary tigernut did not affect total protein and specific acetylcholinesterase activities in the olfactory lobe and medulla regions. However, tigernut intake had significant effects on total protein levels in other brain regions, leading to significant changes in specific acetylcholinesterase levels in the cerebellum. These findings suggest that tigernut added to rabbit diets at levels above 20 g/kg significantly alters brain acetylcholinesterase concentration, total protein levels, and specific acetylcholinesterase, potentially impairing brain functionality.

Keywords: Brain, Tigernut, Rabbit, Genotypes, Acetylcholinesterase, Protein, AChE, SAChE, and Diets.

INTRODUCTION:

The scarcity of animal protein, particularly from animal sources, in the diets of people across Africa and many developing nations is a significant concern (Adedeji *et al.,* 2012). To address this issue, it is crucial to increase the production of non-ruminant species, including rabbits, as they offer a fast and efficient solution for improving animal protein supply in tropical Africa (Adedeji *et al.,* 2012). Despite efforts to meet the demand for animal protein through the consumption of cattle, pigs, poultry, sheep, and goats in Nigeria, the supply remains insufficient (Adedeji *et al.,* 2012). Moreover, the recommended minimum intake of animal protein by the Food and Agriculture Organization (FAO) has not been achieved, largely due to the high cost of meat, which is unaffordable for many Nigerians (FAO, 2011).

Expanding the production of small yet highly prolific livestock is crucial in addressing the low intake of animal protein (Adedeji *et al.,* 2012). Rabbits, in particular, have been identified as an economical livestock option that can help fill the gap in dietary protein intake in Nigeria (Adedeji *et al.,* 2012). These rabbits are micro-livestock, capable of producing approximately 47kg of meat per doe per year, which is sufficient to meet the animal protein requirements of a medium-sized family under small-scale rural farming systems (Adedeji *et al.,* 2012). Furthermore, rabbit meat offers additional nutritional benefits, such as being rich in vitamin B, low in cholesterol and sodium levels, making it an attractive source of high-quality animal protein (Jithendran, 2000; Omole *et al.,* 2005).

The rabbit has gained attention for its potential as an animal that exhibits desirable attributes such as small body size, short generation interval, and high reproductive potential (Adedeji *et al.,* 2012). As an economic source of high-quality animal protein, rabbits are gradually becoming recognized in tropical regions as a means of addressing the nutritional needs of the human population (Adedeji *et al.,* 2012). Known for their prolific nature and efficient conversion of feeds to muscle, rabbits are capable of utilizing diverse forages, which makes them suitable for overcoming the scarcity of cereals in the face of competition from industries and human consumption (Adedeji *et al.,* 2012). To ensure a sustainable balance between population growth and agricultural productivity, it becomes necessary to explore alternative feed options that are lesser known and underutilized (Bamishaiye and Bamishaiye, 2011). One such option is Cyperus esculentus, commonly known as tiger nut, which belongs to the family Cyperaceae (Fasakin *et al.,* 2017; Sharif *et al.,* 2019). Tiger nuts possess essential agronomic and nutritional potentials, comparable to conventionally used energy sources (Fasakin *et al.,* 2017). In Nigeria, there are two main varieties of tiger nut: the yellow and the brown, with the yellow variety being preferred due to its larger size, attractive color, fleshier body, higher milk content, lower fat, and higher protein content, along with fewer antinutritional factors, especially polyphenols (Fasakin *et al.,* 2017). Recent studies have demonstrated that tiger nut residue or the whole plant is rich in energy, fair in amino acid composition, with minimal allergens, making it a valuable ingredient in livestock feed (Ellman, 1961; Reinhold, 1953). However, there is a lack of information regarding the effect of tiger nut meal specifically on rabbits. Hence, this study aims to investigate the impact of varying levels of dietary tiger nut on the acetylcholinesterase (AChE) activities, total protein, and specific acetylcholinesterase (SAChE) of the brain in three different rabbit genotypes.

MATERIALS AND METHODS:

The experiment utilized a total of 320 male and female rabbits, aged 6 weeks, belonging to three different genotypes. The study was conducted for duration of eight weeks at the livestock section of Teaching and Research Farm, The Federal University of Technology, Akure. The rabbits were weighed and randomly allocated to four treatments, comprising diets with tiger nut inclusion levels of 0, 10, 20, and 30g per kg of feed. Each treatment was replicated five times, with 15 rabbits per replicate, following a completely randomized design. After the completion of the experiment, three males and three females from each replicate were slaughtered, resulting in a total of 24 rabbits per treatment. The brains were immediately removed and carefully separated from any attached meninges and blood vessels. Using an ice-cold porcelain tile, the brains were dissected into different regions, including the cerebellum, cerebral cortex, mid brain, and medulla oblongata, following the method described by Adejumo and Egbunike, (2001). The brain and hypophyseal acetylcholinesterase (AChE) activities and total protein concentrations were measured using colorimetric methods as described by Ellmann *et al.* (1961) and Reinhold, (1953) respectively. Randox commercial kits with catalog numbers TP245 and CE7944 were utilized for the determination of total protein (TP) and AChE, respectively. The brain and hypophyseal samples obtained from each animal were homogenized in an ice-cold phosphate buffer solution containing 0.1% Triton X100 using a Potter Elvehjem homogenizer (catalog number P7734) at a concentration of 1% weight per volume (w/v). The specific acetylcholinesterase (SAChE) activity was calculated by dividing the AChE activity of each sample by its corresponding total protein concentration, expressed in µmole/g protein/min. Statistical analysis was performed using SAS (2008, version 9.2), employing One Way Analysis of Variance (ANOVA) with a significance level of α=0.05. In cases where significant differences were found, means were compared using the Duncan Multiple Range Test based on the guidelines outlined by Snedecor and Cochran, (1980).

RESULTS:

The inclusion levels of tigernut did not significantly affect the acetylcholinesterase concentration in the olfactory lobe, cerebellum, and pineal body of the brain (**Table 1**). However, there was a significant effect on the optic lobe and medulla oblongata, where the activity of acetylcholinesterase decreased significantly in the rabbits fed diets containing 10 and 30g tigernut/kg compared to the control and 20g tigernut/kg diet. In terms of breed, there was no significant influence on acetylcholinesterase activity, but sex had a significant effect on the optic lobe, with male rabbits showing higher activity than females. The specific acetylcholinesterase activity in the brain regions (olfactory lobe, pineal body, optic lobe, medulla) did not significantly differ across the varied inclusion levels of dietary tigernut, except in the cerebellum where the activity decreased significantly with increasing inclusion level, with the control group (0g/kg) showing the highest activity (**Table 2**). Breed had a significant influence on the activity in the olfactory lobe, optic lobe, cerebellum, and medulla, except for the pineal body. Among the breeds, Dutch rabbits had the highest activity in the optic lobe, while Hyla max and Newzealand rabbits showed a similar level of significance. Sex did not have a significant influence on the specific acetylcholinesterase activity in the rabbits' brain. Regarding the total protein concentration in specific brain regions (optic lobe, cerebellum, pineal body), there was a significant effect except for the olfactory lobe and medulla (**Table 3**). The pineal body showed an increase in total protein at the 30g/kg inclusion level, similar to the control. The effect of tigernut on the total protein of the optic lobe and cerebellum did not follow a consistent pattern, with the 0g/kg inclusion level group showing the highest total protein. Breed had a significant influence on the optic lobe, with Dutch rabbits recording the highest value, while Hyla max and Newzealand rabbits showed a similar level of significance. Sex had a significant influence on the optic lobe, with male rabbits having a higher total protein concentration than females.

Table 1: Percentage composition of experimental diets fed to matured does and bucks.

Ingredients	Compositions
Maize	8.00
Wheat offal	18.00
Rice bran	42.00
Groundnut cake	5.00
Palm kernel cake	25.50
Limestone	1.00
Grower's Premix	0.10
Salt	0.20
Lysine	0.10
Methionine	0.10
Total	100

Calculated Composition

Metabolisable energy (Kcal/kg)	2586.41
Crude protein	14.86
Crude fibre	13.29
Calcium	0.48
Available phosphorus	0.66
Lysine	1.17
Methionine	0.73

Table 2: Brain Acetylcholinesterase (AChE) Activities of Three Rabbit Breed Fed Different Levels Tiger nut.

Treatment	Olfactory Lobe	Optic Lobe	Cerebellum	Pineal Body	Medulla
10	0.21±0.01	0.16±0.01^{b}	0.14±0.01	0.17±0.01	0.22±0.01^{b}
20	0.21±0.01	0.16±0.01^{b}	0.15±0.01	0.14±0.01	0.25±0.01^{a}
30	0.2±0.01	0.17±0.01^{b}	0.14±0.01	0.17±0.01	0.21±0.01^{b}

Control	0.2±0.01	0.23±0.02^{a}	0.14±0.01	0.15±0.01	0.26±0.01^{a}
Dutch	0.21±0.01	0.19±0.01	0.14±00.1	0.16±0.01	0.24±0.01
Hyla	0.2±0.01	0.17±0.01	0.14±0.01	0.17±0.01	0.25±0.01
Newzealand	0.2±0.01	0.18±0.01	0.14±0.01	0.14±0.01	0.22±0.01
Female	0.21±0.01	0.17±0.01^{b}	0.15±0.01	0.15±0.01	0.23±0.01
Male	0.2±0.01	0.19±0.01^{a}	0.14±0.01	0.16±0.01	0.24±0.01
Treatment	0.40	0.00	0.36	0.08	0.00
Breed	0.88	0.69	0.93	0.05	0.12
Sex	0.67	0.04	0.32	0.44	0.23
Treatment * Breed	0.12	0.60	0.22	0.01	0.02
Treatment * Sex	0.76	0.07	0.00	0.05	0.14
Breed * Sex	0.62	0.15	0.62	0.82	0.18
Treatment * Breed* Sex	0.37	0.28	0.14	0.10	0.00

Values are means ± SEM, Means in a row without a common superscript are significantly ($p<0.05$) different

Table 3: Brain Specific Acetylcholinesterase (SAChE) Activities of Three Rabbits Breed Fed Tiger nut.

Treatment	Olfactory Lobe	Optic Lobe	Cerebellum	Pineal Body	Medulla
10	0.77±0.06	0.91±0.05	0.92±0.05^{bc}	0.94±0.06	0.71±0.08
20	0.74±0.06	0.87±0.05	0.92±0.04^{bc}	1.05±0.08	0.76±0.09
30	0.78±0.03	0.88±0.03	0.82±0.04^{c}	1.02±0.09	0.92±0.1
Control	0.96±0.14	0.91±0.14	1.19±0.08^{a}	1.05±0.07	0.76±0.05
Dutch	0.91±0.1	1.01±0.09	0.99±0.06	0.97±0.05^{b}	0.84±0.08
Hyla	0.76±0.05	0.84±0.05	0.99±0.06	0.95±0.07^{b}	0.74±0.07
Newzealand	0.77±0.05	0.82±0.05	0.92±0.05	1.13±0.07^{a}	0.79±0.06
Female	0.75±0.03	0.85±0.03	0.89±0.03	1.02±0.05	0.8±0.06
Male	0.87±0.08	0.94±0.07	1.04±0.05	1.01±0.06	0.78±0.05
Treatment	0.32	0.95	0.00	0.58	0.23
Breed	0.33	0.07	0.35	0.04	0.50
Sex	0.17	0.32	0.00	0.79	0.79
Treatment * Breed	0.09	0.00	0.06	0.03	0.42
Treatment * Sex	0.10	0.23	0.00	0.00	0.58
Breed * Sex	0.47	0.40	0.01	0.38	0.09
Treatment * Breed* Sex	0.08	0.01	0.20	0.06	0.01

Values are means ± SEM, Means in a row without a common superscript are significantly ($p<0.05$) different

Table 4: Brain Total Protein (g/dL) of rabbits Breeds Fed Different levels of Tiger nut.

Treatment	Olfactory Lobe	Optic Lobe	Cerebellum	Pineal Body	Medulla
10	0.16±0.01	0.14±0.01^{c}	0.13±0.01^{b}	0.15±0.01^{b}	0.15±0.01
20	0.15±0.01	0.14±0.01^{c}	0.13±0.01^{b}	0.14±0.01^{c}	0.19±0.02
30	0.15±0.01	0.15±0.01^{b}	0.12±0.01^{c}	0.16±0.01^{a}	0.18±0.02
Control	0.2±0.04	0.21±0.04^{a}	0.15±0.01^{a}	0.15±0.01^{b}	0.19±0.01
Dutch	0.19±0.02	0.2±0.03^{a}	0.14±0.01	0.15±0.01	0.19±0.01
Hyla	0.15±0.01	0.14±0.01^{b}	0.14±0.01	0.14±0.01	0.17±0.01
Newzealand	0.15±0.01	0.14±0.01^{b}	0.13±0.01	0.15±0.01	0.17±0.01
Female	0.15±0.01	0.14±0.01^{b}	0.13±0.01	0.15±0.01	0.18±0.01
Male	0.18±0.02	0.18±0.02^{a}	0.14±0.01	0.15±0.01	0.18±0.01
Treatment	0.34	0.00	0.00	0.02	0.20

Breed	0.27	0.00	0.54	0.37	0.56
Sex	0.18	0.00	0.06	0.20	0.76
Treatment * Breed	0.09	0.00	0.02	0.00	0.85
Treatment * Sex	0.05	0.00	0.02	0.02	0.13
Breed * Sex	0.26	0.00	0.03	0.88	0.43
Treatment * Breed* Sex	0.03	0.00	0.62	0.77	0.01

Values are means ± SEM, Means in a row without a common superscript are significantly ($p<0.05$) different

Acetylcholinesterase (AChE) levels in the brain of experimental rabbits were highest and significantly different ($P<0.05$) at 0g/kg compared to other inclusion levels. This indicates that inclusion of tigernut above 0 g/kg in rabbit diets leads to a decrease in the sensitivity of AChE to acetylcholine. Similar increases in AChE activity have been reported in previous studies with MSG-treated mice (Lucinei *et al.,* 2000; Sowmya and Sarada, 2015; Khalil and Khedr, 2016; Fasakin *et al.,* 2017). However, the present findings differ from a study by Abu-Taweel, (2016) which reported a decrease in AChE activity in MSG-treated rats. The decreased AChE concentration in the brain of tigernut-fed rabbits may be due to a lack of dysfunction in the cholinergic system, resulting in consistent enzyme activity. The non-significant difference in AChE activity in the brain region of the rabbits may suggest consistent cholinergic neurotransmission efficiency. These findings contradict previous reports that documented varying effects on AChE activity in treated animals. Abu-Taweel, (2016) reported a decrease in AChE activity in MSG-treated rats, while (Lucinei *et al.,* 2000; Khedr, 2016; Fasakin *et al.,* 2017) reported an increase in AChE activity in MSG-treated mice. It is possible that rabbits fed tigernut in excess of 30 g/kg may experience an increase in cholinergic neurotransmission efficiency due to the increased acetylcholine levels caused by decreased AChE activity. The lack of significant effect of dietary tigernut on AChE activity in the olfactory lobes may indicate no impaired sense of smell, in contrast to the assertion of Maynitz, (2018). Additionally, there was no significant difference in AChE activity in the pineal body and cerebellum regions of the rabbits' brains in the treatments containing tigernut. This suggests that the synthesis and catabolism of neurotransmitters (AChE) were not affected by the diet up to 30 g/kg. The insignificant effect observed in the AChE of the pineal body up to the 30 g/kg treatment diet indicates that dietary tigernut at the inclusion levels did not interfere with melatonin production, thereby not negatively affecting the modulation of sleeping patterns and behavior of the rabbits. The results of the brain-specific acetylcholinesterase (SAChE) levels are shown in **Table 4**. A significant influence was observed for the cerebellum, with the control (0g) of tigernut recording the highest SAChE activity. Adejumo *et al.* (2005) did not report any influence of fumonism on SAChE activities in the cerebellum of pigs. The difference in the results may be due to the different species of experimental animals and the different supplements that were fed to them. Adejumo and Egbunike, (2004) agreed with the non-significant influence observed in the SAChE activities of the olfactory lob, optic lobe, pineal body, and medulla oblongata regions of the brain. On the other hand, the total protein concentration in the brains of rabbits fed tigernut at inclusion levels of 0-30 g/kg was depleted compared to the control, except for the pineal body where the 20 g inclusion level recorded the highest concentration. However, the total protein concentration in the olfactory lobe and medulla of the experimental rabbits was not affected. The lack of significant difference in total protein concentration in the olfactory lobes and medulla across all the treatment diets indicates that tigernut had no interference with neural mechanisms involved in protein synthesis in the brain of the rabbits. Conversely, the significant decrease observed in the total protein concentrations in the other regions of the brain at higher dosages of tigernut (10 to 30 g/kg) suggests that tigernut disrupts protein synthesis in those regions. Ewuola and Bolarinwa, (2017) explained that the ability of aflatoxin to bind and interfere with enzymes and substrates involved in protein synthesis makes it capable of affecting brain development. If there were a significant difference in total protein in the olfactory lobe, this could have led to retina dysfunction in the rabbits. Protein in the brain functions for repair of worn-out tissues, growth, muscular development, and also binds to some minerals to ensure their bioavailability and proper utilization (Ewuola and Bolarinwa, 2017).

CONCLUSION:

This study explored the potential of tigernut as a dietary component for rabbits. The findings suggest a two-sided effect: It enhances flavor and provides valuable energy (with safe levels at 10-20g/kg diet) due to its carbohydrate, fats and fiber composition. However, high intake appears to be toxic to neurons in the brain, potentially affecting cognitive function. Further research is needed to understand why and determine the safe limit. The study also suggests excessive tigernut disrupts internal balance (homeostasis). More research is needed to pinpoint the specific effects. Despite its taste and energy benefits, caution is needed regarding its neurotoxic effects. Unraveling the impact of tigernut on rabbits' brain functions and overall health is crucial to determine safe levels for this intriguing ingredient. This knowledge would significantly improve animal nutrition, making this a novel area for further research.

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CONFLICTS OF INTEREST:

The authors declare that they have no conflicts of interest.

REFERENCES:

1) Abu-Taweel, G.M. (2016). Effect of monosodium glutamate and aspartame on behavioral and biochemical parameters of male albino mice. *African J. of Biotechnology,* **15**(15), 601-612. https://www.ajol.info/index.php/ajb/article/view/134376
2) Adedeji, I. A., Adejumo, I. O. & Obaniyi, K.S. (2012). Information needs of farmers in rabbit production in sagamu Local Government Area of Ogun State. *Continental J. of Agricultural Economics*, **6**(1), 23-27.
3) Adejumo DO, Egbunike GN. (2001). Effect of prepubertal, pubertal and postpuberta-lorchidectomy and testosterone therapy on the-acetylcholinesterase activity and protein content in brain and hypophyses of Pigs. Trop. *Anim. Prod. & Inv. J.*, **4**, 157-165.
4) Adejumo, D.O. and Egbunike, G.N. (2004). Changes in acetylcholinesterase activities in the developing and aging pig brain and hypophyses. *Inter J. of Agricultural and Rural Development.* **5**, 46 -53.
5) Adejumo, D.O., Ladokun, A.O., and Sokunbi, O.A. (2005). Species differences in acetylcholesterase activity and total protein concentration in the brain and hypophyes of red sokoto goats and gudalicattles. *Asset Series A*., **5**(1), 121- 127.
6) Ellman, G.L., Courtney, K.D., and Featherstone, R.M. (1961). A new and rapid calorimetric determination of acetylcholinesterase activity. *Biochemical Pharmacology*. 7, 88 -95.
7) Ewuola, E.O., and Bolarinwa, O.A. (2017). Acetylcholinesterase, glucose and total protein concentration in the brain regions of West African dwarf goats fed dietary aflatoxin. *J. Veterinary Medical & Animal Health*. **9**(9), 240-245.
8) FAO, (2011). Food outlook global market analysis. Rome Italy: Food and Agricultural organization Jackson, R. A.
9) Fasakin, O.W., Fajobi, A.O., and Oyedapo, O.O. (2017). Neuroprotective potential of Aframomummelegueta extracts on brain of monosodium glutamate-treated wistar albino rats. *J. of Neuroscience and Behavioral Health*, **9**(2), 16-27.
10) Jithendran, K. P. (2000). Nutritive value of rabbit meat. Envis Bulletin Himalayan Ecology and Development, **8**(1).
11) Khalil, R. M., and Khedr, N.F. (2016). Curcumin protects against monosodium glutamate neurotoxicity and decreasing NMDA2B and mGluR5 expression in rat hippocampus. *Neurosignals*. **24**, 81-87.
12) Lucinei, B.S., Gravena, C., and de Freitas Mathias, P.C. (2000). Insulin Secretion and Acetylcholinesterase Activity in Monosodium L-Glutamate-Induced Obese Mice. *Hormone Research. In Pediatrics*. **54**(4), 1186-1191. https://doi.org/10.1159/000053257

13) Maynitz, M. (2018*)*. Bird senses - sight, hearing, touch, taste and smell. How birds use their sense. The spruce. https://www.thespruce.com/birds-fivesenses-386441
14) Reinhold, J.G. (1953). Manual determination of serum total protein, albumin and globulin fraction by buiret method In: Reiner, M. (Ed). Standard methods in clinical chemistry. *Academic Press, New York,* Pp: 88 – 97.
15) Sharif IH, Tamanna S, Mosaib MG, Tamanna N, Haque MA, Jamal MAHM, and Uddin ME. (2019). Assessment and biomonitoring of the effect of rapeseeds oil on wister rat organs. Am. *J. Pure Appl. Sci*., 1(4), 20-29. https://doi.org/10.34104/ajpab.019.0192029
16) SAS Institute Inc. (2008). SAS / STAT User's Guide. Version 9.2 for Windows. SAS Institute Inc; SAS Campus Drive. Carry, North Carolina, U.S.A.
17) Snedecor GW, and Cochran WG. (1980). Statistical methods. Book, PP. 4207th ED. Lowa Stat. *Univ. Press*, Ames, Lowa , U.S.A
18) Sowmya, M. and Sarada, S. (2015). Combination of Spirulina with glycyrrhizin prevents cognitive dysfunction in aged obese rats. *Indian J. of Pharmacology*. **47**(1).
19) Windisch, W. M., Schedle, K., and Kroismayr. (2008). Use of phytogenic products as feed additives for swine and poultry. *J. of Animal Science*. **86**, 140-148.

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