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Original Research Article

Allicin Improves Olfactory Response to Rewarding Stimuli Using Buried Reward Test

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Abstract: Garlic, known as *Allium sativum*, is used in food and medicine. Allicin is one of the main bioactive agents found in garlic. This study investigated the tendency for allicin, a bioactive agent in garlic, to improve olfactory response in male wistar rats exposed to cadmium chloride-induced toxicity. Thirty male wistar rats were sampled in six groups of five rats each. The treatments were; group 1 (control)- water and feed, group 2-$CdCl_2$ (5mg/kg) group 3-allicin(20mg/k), group 4-allicin(40mg/kg), group 5- $CdCl_2$ (5mg/kg) +allicin (20mg/kg), group 6- $CdCl_2$ (5mg/kg) +allicin(40mg/kg). Olfactory response was tested using buried reward test with the result presented as latency (in seconds). Statistical significance was tested at 95% confidence interval ($P \leq 0.05$). There was a significant decrease ($P \leq 0.05$) in latency and an increase in olfactory response in response to increase in the dose of allicin administered. $CdCl_2$ alone caused a significant increase ($P \leq 0.05$) in latency and decrease in olfactory response but when combined with allicin treatment there was a significant improvement in olfactory response. Allicin improves olfactory response dose-dependently using buried reward test.

Keywords: Allicin; Olfaction; Toxicity; Latency; Response.

INTRODUCTION

Allicin is a bioactive molecule from garlic (*Allium sativum*) produced from an organic compound called alliin when fresh garlic is crushed or cut [1, 2]. Allicin is an Allyl compound which are organic molecules found in garlic and other vegetables of the *Allium* genus [2, 3]. These allyl compounds include allicin, alliin, S-allycysteine, diallyl sulfide, diallyl disulfide and S-allylmercaptocysteine [4]. When garlic is ingested, the allicin it contains is usually degraded to reactive sulfur-containing compounds. Allicin has both agricultural and medicinal uses [4, 5]. In agriculture, allicin can be used in plant protection against many pathogenic fungi such as *Alternaria brassicicola*, *Botrytis cinerea*, *Magnaporthe grisea*, *Plectospherella cucumerina* and *Xanthomonas grisea* [5, 6]. In medicine, allicin is known to be very reactive and an antioxidant that maintains membrane stability [7]. Allicin and diallyl disulfides account for the pungency and spicy aroma of fresh garlic [5-8]. Olfaction is widely conserved in evolution and allows animals to collect information in their environment. For most animal species, olfaction plays a paramount role in their perception of the environment in terms of the possibility to assess social relationships or finding a sexual partner or food and to detect danger. Lower animals like wistar rats have efficient olfactory discriminatory and cognitive abilities [9]. Diseases as well as some environmental factors like exposure to toxins, radiation and stress may affect olfaction and overall behavior in wistar rats [10]. Despite the pungent nature of garlic, there is scarcity of study investigating the effect of its bioactive agent, allicin, on olfactory behavior in normal and toxicity-induced conditions. This study determined the influence of allicin in olfactory behavior in response to a rewarding stimulus using a buried reward test.

METHODS

Ethical approval

This study followed the guidelines for animal use in experimental research according to the Research Ethics Committee of University of Port Harcourt, Choba, Port Harcourt, Nigeria, with reference no. UPH/CEREMAD/REC/MM70/40.

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Toxicity study

The oral LD_{50} of allicin for male wistar rats is 309mg/kg [4]

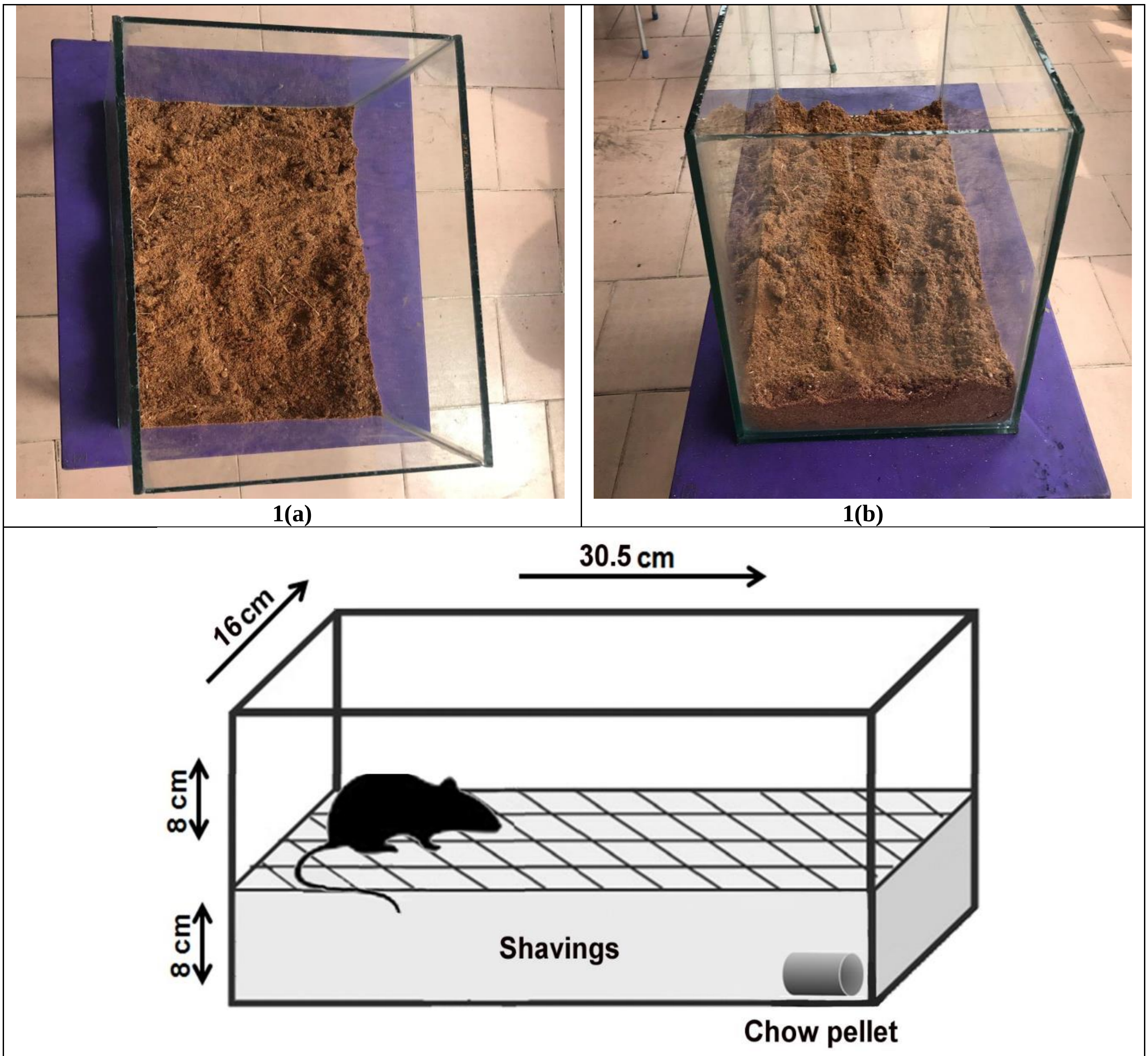
The oral LD_{50} of $CdCl_2$ for male wistar rats is 88mg/kg [10]

Study Design

This study sampled thirty (30) male wistar rats aged 5 weeks old, weighing 140 to 150 grams. The animals were housed in six (6) separate steel cages measuring 59.8x38x22 in centimeters (cm). Each cage contained 5 rats. After 2 weeks acclimatization, the study lasted for 6 weeks. The groups and their oral administrations are as follows; group 1 (control)- water and feed, group 2-$CdCl_2$ (5mg/kg) group 3-allicin(20mg/k), group 4-allicin(40mg/kg), group 5-$CdCl_2$ (5mg/kg) +allicin(20mg/kg), group 6- $CdCl_2$ (5mg/kg) +allicin(40mg/kg).

Test for olfactory response

Olfactory response was tested using the buried reward test. A wistar rat is placed in a cage bedded with saw dust that measures 30.5x16x16 cm. in an 8cm thick bedding, the reward stimulus (chow pellet) was buried 7cm into the bedding such that it cannot be seen from the surface. the time taken for the animal to dig and sniff the chow pellet was recorded. The maximum time, recorded as latent period, was 10 minutes (600 seconds).



1(c)

Figures 1 (a); Top view of a buried reward chamber, 1(b); Side view of a buried reward chamber, 1(c); Dimensions of a buried reward chamber

RESULTS

The results of this study is presented as follows:

Table 1: Total antioxidant capacity of allicin using ascorbic acid as standard

Concentration (mg/ml)	Ascorbic acid	Allicin
20	0.140	0.097
40	0.176	0.128
60	0.211	0.142
80	0.268	0.186
100	0.322	0.227

From Table 1 showing the total antioxidant capacity (TAC) of allicin using ascorbic acid as standard, with an increase in concentration (in mg/ml) of allicin, from 20mg/ml to 100mg/ml, there was a corresponding increase in TAC. Ascorbic acid showed a greater TAC compared to allicin in same doses.

Table 2: Effect of allicin on olfactory response before the onset of administration of any agent (day 0)

Groups	Latency (sec.)	% Change
Control	304.1±1.12	0
$CdCl_2$ (5 mg/kg)	304.4±1.13	0.09
Allicin (20mg/kg)	307.2±1.33	1.01
Allicin (40mg/kg)	303.4±0.42	-0.23
$CdCl_2$ (5mg/kg)+allicin (20mg/kg)	303.2±1.13	-0.29
$CdCl_2$ (5mg/kg)+allicin (40mg/kg)	305.3±1.10	0.39
N=5		

From Table 2 showing the effect of allicin on olfactory response before the onset of administration of any agent (termed day 0 of the experimental period), the olfactory response in all test groups was similar and showed no significant difference ($P \leq 0.05$). For all test groups relative to control, the percentage change (% change) was slightly above 1.

Table 3: Effect of allicin on olfactory response after administration (day 42)

Groups	Latency (sec.)	% Change
Control	300.1±1.12	0
$CdCl_2$ (5 mg/kg)	520.4±1.17*	73.40
Allicin (20mg/kg)	251.2±1.33*	-16.29
Allicin (40mg/kg)	170.3±0.41*	-43.25
$CdCl_2$ (5mg/kg)+allicin (20mg/kg)	380.1±1.20*	26.65
$CdCl_2$ (5mg/kg)+allicin (40mg/kg)	280.3±0.11*	-6.59
N=5		

From Table 3 showing the effect of allicin on olfactory response after administration (day 42 of experimental period) there was a progressive significant increase ($P \leq 0.05$) in olfactory response that positively correlated with the dose of allicin administered independently or in combination with $CdCl_2$. The highest percentage change relative to control was observed in $CdCl_2$ 5mg/kg independent treatment which presented a delayed or prolonged olfactory response. Allicin 40mg/kg treatment presented the fastest latency compared to the other groups.

DISCUSSION

This study investigated the tendency for allicin, a bioactive agent in garlic, to improve olfactory response in male wistar rats. It is a dose dependent study that assessed the response of wistar rats to a buried rewarding stimulus. Olfaction is a nervous response that involves alteration of neural signals [11]. Allicin potentiates several neural responses even though literature explaining its effect on olfaction is scarce [11, 12]. From this study, as the dose of allicin administered was increased, there was a progressive shortening of latency. The shortened latency is translated as an increase in olfactory response [13]. However, allicin may have altered latency may be at musculoskeletal or neuromuscular or both levels. The antioxidant effect of allicin may be beneficial in opposing the toxicity of cadmium chloride and its tendency to adversely affect olfaction [5]. Allicin slows down the death of neurons and reduced cognitive deficits in Alzheimer's disease patients [14]. Cadmium chloride is a neurotoxic agent that has been reported to damage central and peripheral nervous system neurons. The combination of both allicin and cadmium chloride may have reversed or prevented the damaging effect of cadmium chloride on olfactory neurons. The mechanism by which cadmium chloride manifests its neurotoxicity is believed to be through generation of free radicals at cellular level and subsequent peroxidation and oxidative modification of biomolecules [15, 16]. The antioxidant effect of allicin may have been effective enough to combat free radical generation in neurons that may impair olfactory response in wistar rats. Allicin protects spinal cord neurons from oxidative modifications induced by heavy metals [5].

CONCLUSION

This study showed that there is a tendency for allicin to improve olfactory response both in normal and neurotoxicity-induced conditions in male wistar rats. The effect allicin may have in both conditions may be dose-dependent.

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