IN VITRO EVALUATION OF ANTI-CONVULSANT POTENTIAL OF ALLIUM SATIVUM EXTRACT



Pydiraju Kondrapu¹, Shailaja Nare², Ganesh Rangrao Pawar^{3*}, A. Rekha Devi⁴, Mamta Bhatia⁵, Nisha Choudhary⁶, Talele Dipali⁷, Moushumi Baidya⁸

Article History: Received: 11.04.2023	Revised: 27.05.2023	Accepted: 13.07.2023

Abstract

Allium sativum, also known as ALLIUM SATIVUM, is a basic vegetable that has traditionally been used for cooking, flavoring, and natural remedies. Patented organic sulfur compounds in ALLIUM SATIVUM include diallyl sulfide, allicin (dially lthiosulfate), -glutamylcysteine, S-allylcysteine (alliin), and ajoene. ALLIUM SATIVUM positively affects stimulation, oxidative pressure markers, hypertension, hyperlipidemia, and endothelial capacity in vitro or in animal models. In addition to their use in humans, these bioactive atoms play a significant role in the creation of domesticated animals and fish. The modern rural concept of natural animal culture is dependable with the addition of ALLIUM SATIVUM and its related goods to animal feed. This study collects information on the effects of using ALLIUM SATIVUM and its extracts on certain animal execution limits, including chicken, hares, ruminants, pigs, and fish. This audit may serve as a guide for researchers and businesspeople as they investigate the uses of feeds containing ALLIUM SATIVUM and allicin side effects to enhance animal husbandry and seafood output.

Keywords: Animal production, allium sativum, Nutritional applications, anti-convulsant potential, allium sativum, extract.

¹Assistant Professor, Aditya pharmacy colleg, ADB road , Surampalem, Kakinada, Andhra Pradesh. 533437

²Assistant professor, Bharati Vidyapeeth College of Pharmacy, Near Chitranagari, Kolhapur, Maharshtra- 416013

^{3*}Department of Pharmacology, NSPM'S College of Pharmacy, Arni Bypass Road Darwha District Yavatmal, Maharashtra, India. 445202

⁴Associate Professor, SEVEN HILLS College of Pharmacy, Venkataramapuram, Tirupati, Andhra Pradesh. 517526

^{5,6}Assistant Professor, Maulana Azad University, Bhujawar, Jodhpur, Rajasthan. 342802

⁷Assistant Professor, Faculty of Pharmacy, Vishwakarma University, Survey No 2,3,4 Laxmi Nagar, Kondhwa, Budruk, Pune. 411048

⁸Assistant Professor, Bharat Pharmaceutical Technology, Amtali, Agartala, Tripura. 799130

*Corresponding Author

Ganesh Rangrao Pawar^{3*} ^{3*}Assistant Professor, NSPM'S College of Pharmacy, Arni Road Darwha Dist Yavatmal, Maharashtra, India. 445202

DOI: 10.31838/ecb/2023.12.s3.672

1. Introduction

Over best the past 100 vears. the biotechnology improve livestock to productivity has been the use of antibiotic growth promoters (AGPs) in nursing. AGP offers a wide range of skills, including those that have been used for many years in hydroponics and animal husbandry, such as disease prevention and treatment. [1] However, misuse of AGPs leads to drug deposition and bacterial blockage, severely

affecting livestock and seafood quality, and ultimately threatening safe food handling and human health. However, long-term use of antibiotics has raised concerns about bacterial blockage and drug accumulation, leading to increased restrictions on the use of antibiotics in pig farming. Numerous studies have shown that common growth promoters such as activating creams, probiotics, chitooligosaccharides and other additives can replace AGP in animal nutrition without affecting animal performance shown.[2]



Figure 1:the ability of allium sativum extract to prevent seizures

One of the most well-known natural remedies is the edible plant Allium sativum, which has long been regarded as a medical marvel of great interest throughout human history. Allicin, a non-protein amino caustic, has been identified as a major component of leeks by phytochemical analysis. It is cleaved to allicin by alliinase (aliin lyase, EC 4.4.1.4, α, βscavenging endogenous lyase from Allium) and many other sulfur compounds (such as diallyl sulfide and γ-glutamyl-S-). Configurable. Allyl-L-cysteine and S-allyl-Lcysteine sulfoxide (alliin) and ajoene,

commonly referred to as allium compounds. [3] The contained organ Sulfur compounds in allium sativum, together with the plant's distinct pungent scent and other therapeutic characteristics, are what give it its powerful pharmacological effects.

As a result, the purpose of this research is to demonstrate how Allium sativum and Allicin can take the place of AGP in animal husbandry. This article reviews the use of leeks and their primary extract, allicin, in domestic and marine animals such as chickens, pigs, and fish.

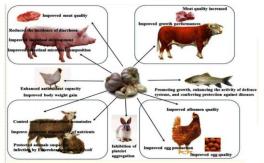


Figure 2: The extraordinary advantages of Allium sativum in animals.

Literature review

Lang et al (2004) thought about how allicin affects gastrointestinal epithelial cells in terms of immunomodulation. HT-29 and Caeo-2

cells' unrestricted and TNF-activated release of IL-1, IL-8, IP-10, and MIG were examined without allicin pretreatment. Utilizing ELISA,

cytokinine emission was assessed, and nonbinding RNA assurance was determined.

Betaneor et al (2003)evaluated the Trolox equivalent antioxidant activity (TEAC) of Allium sativum to determine its antioxidant capacity. [4] They demonstrated that copper and the absolute phenolic compound both have significant antioxidant action.

Fuhrman et el (2000)focused on the synergistic antioxidant effects of allium sativum with other common antioxidants. They oversaw a review in this situation to determine the effect of tomato lycopene alone or in combination with other common antioxidants found in allium sativum on LDL oxidation. [5] The phenolics rosmarinic and eamosic, as well as the flavanoidsglabridin and allium sativum, were thought to partially inhibit LDL oxidation.

Morihara et al (2006) focused on the effect of matured allium sativum extract (Progress in years) on rodent weakness. Rodents were subjected to persistence training on a mechanical treadmill device many times each week for a substantial amount of time. 30 minutes before to each exercise, animals were given AGE at a dose of 2.86g/kg. In the gastrocnemius and soleus muscles, succinate dehydrogenase (SDH), superoxide dismutase (SOD), metabolites of nitric oxide (NO), and lactic corrosive cone.

2. Materials and Methods

Collection of plant materials

New A. sativum and A. cepa were bought from the Ede market in Osun State, Nigeria and delivered to the Organization of Microbiological Sciences, Adeleke College, Ede. Oyawoy, O.M. officially recognized plant models by looking at their actual qualities. These samples don't have any vouchers attached to them.

Preparation of plant extract

The raw plants of A. sativum and A. cepa were chopped, ground, and then burned to dry into powder. Shuford et al. state that, 100 g of squashed green onions and onions were submerged in 1000 ml of decontaminated water and 95% ethanol answer for separate Chen et al (2007)Allium sativum oil supplementation (rich in diallyl trisulfide) was seen to delay draining 27-time, lengthen thrombin time, and improve anticoagulant factor activity in rats at 5 and 50 mg/kg body weight doses, respectively. [6] This is as a result of the DAT-rich allium sativum oil's anticoagulant properties. However, high allium sativum oil intake substantially increased plasma fibrinogen focus and certain parameters, hematological including erythrocyte count, hemoglobin, and platelets.

Takasu et al (2006)Considering that oxidative characteristics play a significant role in the problem of sickle-cell fragility, the antioxidant effect of matured allium sativum extract (Progress in years) on sickle red platelets was evaluated. Five individuals with sickle cell anemia received AGE at a dose of 5 ml per day. For Heinz body assessment, complete blood tests were collected at pattern and at approximately a month.

Demeule et al (2004)p-glycoprotein (p-gp) and multidrug safe protein 2 (Mrp2) in the renal brush line layer, which were carriers and involved with the guard of cell and multidrug opposition, were the focus of two organosulfur compounds (OSCs), Fathers and SAC. They demonstrated that fathers start Mrp2 expression by two folds, increasing levels of glutathione Stransferase (GST) and glutathione forms.[7] (GSH)-

physiologically dynamic substances from the green onions. To forestall dissipation, the blend was set in a carafe enveloped by clean aluminum foil and kept at room temperature for 48 h [8]. Using Whatman #1 channel paper, the scattered green onions (A. sativum) and onion (A. cepa) were separated to produce a filtrate. A rotary evaporator was used to evaporate the extract. For biochemical and microbiological research, pools were dried on cardboard, and filtrates were extracted and stored in the refrigerator.

Preparation of extracts serial dilution

In a clean test tube, three successive weakenings of each extract were prepared. Aseptically collected ethanol groups and liquid extracts of A. cepa and A. sativum at concentrations of 50, 100, and 150 mg/mL

were utilized. The extract was diluted to a 50 mg/ml attenuation standard by adding 0.5 g to 10 ml of sterile purified water, 100 mg/ml by adding 1 g, and 1.5 g by 10 ml of sterile purified water. was added, resulting in 150 mg/ml.

Collection of bacteria samples

The living range of the Department of Clinical Parasitology and Microbiology, University Emergency Clinic, Ibadan, Oyo provided the organic test materials for this investigation. In this study, Staphylococcus epidermidis, Proteus vulgaris, Pseudomonas aeruginosa, Escherichia coli, and Klebsiella pneumoniae were used as microorganisms. Organic test organisms were streaked onto slanted plates containing additional agar and first incubated at 37°C. A way of life practiced in a cold environment.[9]

Screening and determination of total phenolic compound

By bubbling 2 grams of the test material with 50 milliliters of diethyl ether for 15 minutes in a water shower, anhydrous phenol is extracted. Calorimeter readings are used to complete the evaluation. In a clean laminar flow hood, mix 1 mL of the test solution with 1 mL of the Folin-Ciocalteus phenol reagent for 15 minutes. After cooling, place the mixture on a Petri dish. Each plate has 6 mm suction wells and the plate is inoculated with 30 µl of inoculum. Following that, each test will be pipetted into each well at a concentration of 1000 g/75 l, and the plates will then be incubated for 24 hours at 37 °C. Ones of constraint will be calculated in millimeters (mm) after 24 hours.

Determination of diameter (mm) of zone of inhibition

As per Onyeagba et al., After being thoroughly washed, the inoculum solution was dried for five minutes on 20 milliliters of microbial supplemental agar before becoming solidified. Using a micropipette, a hole 6 mm in diameter was drilled into the growth agar medium. For proper distribution, his 0.5 milliliter aliquots of each plant's unpurified extracts (50, 100, or 150 mg/ml) were poured into each culture medium well. The wells were then incubated for a total of 24 hours at 37°C. The dimensions of the containment zone below are determined in millimeters (mm).

3. **Results and Discussion**

The entire phenolic content of A. cepa and A. sativum is depicted in Figure 1, Figure 2, and Table 1. Table 1 shows the breakdown of the phenolic parts of A. Utilizing Cepa (onion) and A. sativum (Allium sativum) for both and ethanol separates. Phenol water concentrations were lowest in ethanol extracts of A. [10] sativum and highest in liquid extracts of A. cepa. Allium sativum, also known as red, brown, and white onions, all contained varying amounts of phytochemicals. As depicted. As can be seen in Figures 1 and 2, the phenol content of the liquid extracts of A. cepa (0.5550 0.36) and A. sativum (0.254 0.21) was significantly higher than that of the corresponding ethanol extracts (0.1520 0.15), and the phenol content of each species (0.1322)0.010) was significantly higher than that of the other species.

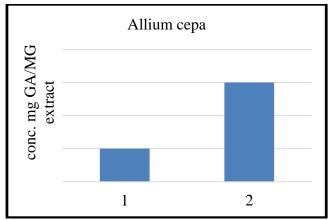


Figure 3: The A. cepa's total phenolic concentration

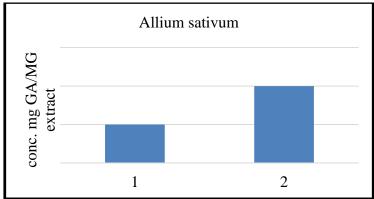


Figure 4:The amount of phenols in the A. sativum plant overall

Table 1:Complete phenolic content of Allium sativum (A. sativum) and A. cepa (onions)

Plants samples	Concentration (mg of GA/mg of extract)
Allium cepa aqueous extract	1.4441 ± 1.25
Allium cepa ethanol extracts	1.0411 ± 1.04
Allium sativum ethanol extracts	1.0211 ± 1.01
Allium sativum aqueous extract	1.1432 ± 1.10

Table 1 shows the total phenol content of Allium sativum (A. sativum) and A. cepa (A. cepa) measured from different concentrations by Onyeagba et al. Confirmed. The antimicrobial efficacy of various concentrations of A. Table 2 displays the effectiveness of the water/ethanol extract of sativum against Proteus mirabilis, Klebsiella pneumoniae, Pseudomonas aeruginosa,

Staphylococcus aureus, and Escherichia coli. The organic restricted zone ranged from 3.0 to 21.4 mm, with S. aureus having the highest retention zone (21.4 mm at 150 mg/mL of the ethanolic extract). Pseudomonas aeruginosa strains in 150 mg/ml aqueous extracts and Klebsiella pneumoniae strains in 100 mg/ml ethanolic extracts showed the least restriction (3 mm). [11]

	Diameter of inhibition zone (mm) of different concentration					
Test bacteria	150 mg/ml		100 mg/ml		50 mg/ml	
	Aqueous	Ethanol	Aqueous	Ethanol	Aqueous	Ethanol
1. Escherichia coli	8	5.1	1	4.1	1	1
2. Staphylococcus aureus	4.4	21.4	1	5.1	1	1
3. Pseudomonas aeruginosa	2.1	5.1	1	4.8	1	1

4. Klebsiella pneumonia	4.1	3.5	1	2.1	1	1
5. Proteus mirabilis	3.1	4.4	1	4.4	1	1

The antimicrobial viability of different groupings of A. cepa water/ethanol separate against Proteus mirabilis, Klebsiella pneumoniae, Pseudomonas aeruginosa, Staphylococcus aureus and Escherichia coli is displayed in Table 3. Biodisorder region expanded from 4.0mm to 25mm. Klebsiella pneumoniae had the lowest restriction (100 mg/mL for liquid extract) while S. aureus had the highest restriction (25 mm at 150 mg/mL for ethanolic extract).

Table 3:Different aqueous/ethanol A. cepa extract concentrations have antibacterial action

Test bacteria	Diameter of inhibition zone (mm) of different concentration					
	150 mg/ml		100 mg/ml		50 mg/ml	
	Aqueous	Ethanol	Aqueous	Ethanol	Aqueous	Ethanol
1. Escherichia coli	4.6	21	3.1	7	1	1
2. Staphylococcus aureus	4.1	25	3.1	5	1	1
3. Pseudomonas aeruginosa	4.1	8	2.4	5	1	1
4. Klebsiella pneumonia	3.4	1	2.1	1	1	1
5. Proteus mirabilis	1	5	3	3	1	1

Phenol content was highest in aqueous extracts of A. cepa and lowest in ethanolic extracts of A. sativum. Red, brown and white onions, Allium sativum, contained different levels of phytochemicals. This supported similar reflective studies conducted by Gazuwa et al. on onion and Allium sativum. Additionally, Micová et al. To support total polyphenol concentration, absolute sulfur content, and antioxidant activity, we focused on Allium. [12] The plant's potential as an antioxidant and tonic is also demonstrated by the extracts' high phenolic content. Allium sativum's health benefits are dependent on its bioactive components, particularly phenolic compounds, which are present relatively in high concentrations and have distinct pharmacological effects. Phenolic compounds are massive collections of optional metabolites that have the power to kill or put out the flames of the free revolutionaries. The

majority of polyphenols discovered in plants are flavonoids and their offshoots. The content of naturally dynamic mixes (polyphenols, sulfur mixtures, and antioxidant dynamic fixing), which varies across cultivars, is what gives allium sativum its medicinal benefits. Ren et al. studied the polyphenol content and antioxidant activity of two onion cultivars cultivated under natural and conventional production conditions and found that the values of total phenol content and total flavonoid content differed significantly from year to year and under natural production conditions. and found to be fundamentally different under conventional production conditions. Natural growing processes have resulted in much higher concentrations of potentially beneficial compounds in onions. Benkebria is testing the antibacterial activity of different phenolic combinations from three different onion species and Allium sativum

against three plants: Aspergillus niger, cyclopium, and Fusarium Penicillium oxysporum. These microorganisms include Salmonella enteritidis and Staphylococcus aureus. The leek showed the least impairment and the leek the most severe impairment. Fusarium oxysporum has the highest consciousness. Antibacterial activity of extracts from A. [13] The sativum species found in this study are consistent with species of Zachariah and Astar. They focused on the effect of leek extract on specific Gram-positive and Gram-negative microorganisms in terms antimicrobial properties. The most of pronounced inhibitory effect against all tested bacteria was found in a study by Shobana et al. Zakaria and Astal also reported that a novel leek extract could inhibit the development of Staphylococcus aureus, Typhoid, and E. coli. coliform bacteria. In a study by Enejiyon et al., a CH3)2CO extract of Klebsiella pneumoniae showed no antibacterial activity in any of the tested populations, consistent with the lowest resistance to Klebsiella pneumoniae. The most likely test separation is actually he correlated with Enejiyon et al. and is Staphylococcus aureus. Although not proven, factors for these organic entities' high impotence were hypothesized to include the existence of phytochemical components. The best extraction solvent was ethanol, perhaps due to the greater ease with which active synthetic compounds dissolved in alcohol. As a result, Staphylococcus aureus was more sensitive to Aspergillus sativum extract than E. coli, which is most susceptible to Aspergillus cepa extract. Nevertheless, the high fragility of these species is said to be due to the plant constituents contained in the plant extracts. Due to their weak inhibitory effects, the this used investigation extracts in demonstrated that Allium species had a significant influence on pathogenic microbes. Both A. sativum and A. cepa had rather modest inhibitory fixations, indicating that their bioactive components had the strongest antibacterial properties.

4. Conclusion

Allium sativum and its extracts have been thoroughly examined by researchers both domestically and internationally, with a focus on their uses in both animal production and human therapeutic trials. However, more research has to be done on the aspect of some of its pharmacological effects.[14] Allicin is an organically active chemical that is taken from the allium sativum bulb and may also be added synthetically. Currently, the extraction rate from fresh leeks is about 0.3%, while the compound allicin has a prediction rate of 85% to 90%. Therefore, synthetic allicin is widely used in animal production due to its low cost, excellent purity of active ingredients and great therapeutic benefits. Antibiotics cannot currently be replaced by allicin, but with the development of modern science and innovation, allicin and other additional substances may be able to do so, leading to the production of pollution-free meat, improved government support for animals, it may contribute to the production of manageable animals. advances in animal husbandry.[15]

5. References

- 1. Abdel-Daim, Abdelkhalek& Hassan (2015) Abdel-Daim MM, Abdelkhalek NK, Hassan AM. Antagonistic activity of dietary allicin against deltamethrininduced oxidative damage in freshwater Nile tilapia: Oreochromis niloticus. Ecotoxicology and Environmental Safety. 2015; 111:146– 152.
- 2. Abdulaziz, Khan & Rahi (2016) Abdulaziz AA, Khan AA, Rahi MJ. Effects of adding different dietary levels of ALLIUM SATIVUM (Allium sativum) powder on productive performance and egg quality of laying hens. International Journal of Poultry Science. 2016; 15:155.
- 3. Betancor-Femandez A, Perez -Galvez A, Sies H, Stahl W. Screening pharmaceutical preparations containing extract turmeric rhizome, artichoke leaf , devil's claw root and ALLIUM SATIVUM salmon oil for antioxidant capacity. J. Pharm Pharmacol. 2003 July; 55(7): 98-6.
- Ameh GI, Eze SC, Omeje FU (2012) Phytochemical screening and antimicrobial studies on the methanolic bulb extract of Allium sativum L. Afr J Biotech 12(14):1665–1668

- Fuhrman B, Volkvo N, Rosenblat M, Aviram M. Lycopene synergistically inhibits LDL oxidation in combination with vitamin E,glabridin, rosmarinic acid , carnosic acid or ALLIUM SATIVUM . Antioxid Redox Signal. 2000 Fall; 2 (3): 491-506.
- Chan KC, Yin MC, Chao WJ. Effect of diallyl trisulfide—rich ALLIUM SATIVUM oil on blood coagulation and plasma activity of anticoagulation factors in rats. Food Chem Toxicol. 2007 Mar! 45 (3): 502-7
- Demeule M, Brossard M, Tureott S, Regina A, Jodoin J, Beliveaur R. Diallyl disulfide ,a chemoprotective agent in ALLIUM SATIVUM .induces multidrug resistance associated protein 2 expression . BiochemBiophyl Res Commun.2004 Nov 12; 324(2): 937-45.
- Amin et al. (2015) Amin AB, Yousuf MB, Kolo UM, Ibrahim AA, Muhammad AI, Maigado AI. Influence of periodic administration of ALLIUM SATIVUM extract on blood parameters of Grazing Lambs. Biokemistri. 2015;26(4):114– 119.
- 9. Anyawu MU, Okoye RC (2017) Antimicrobial activity of Nigerian medicinal plants. J IntercultEthnopharmacol 6(2):240–259
- 10. Ao et al. (2010) Ao X, Yoo JS, Lee JH, Jang HD, Wang JP, Zhou TX, Kim IH. Effects of fermented ALLIUM

SATIVUM powder on production performance, egg quality, blood profiles and fatty acids composition of egg yolk in laying hens. Asian-Australasian Journal of Animal Sciences. 2010;23(6):786–791.

- Bakht J, Khan S, Shafi M (2013) Antimicrobial potential of fresh Allium cepa against gram positive and gram negative bacteria and fungi. Pak J Bot 45(1):1–6
- 12. Benkeblia N (2005) Antimicrobial activity of essential oil extracts of various Onions (Allim cepa) and ALLIUM SATIVUM (Allium sativum). Lebensm-Wiss-u-Technol 37:263–268
- Lang A, Lahav M, Sakhnini E, Barshack I, Fidder HH, Avidan B, Bardan E, Hershkoviz R, Bar- Meir S, Chowers Y. Allicin inhibits spontaneous and TNFalpha induced secration of proinflammatory cytokines and chemokines from intestinal epithelial cells. Clin Nutr. 2004; 23 (5): 1199- 208.
- Morihara N, Ushihama M, Kashimoto N, Sumioko I, Nishihama T, Hayama M, Takeda H. Aged ALLIUM SATIVUM extract ameliorates physical fatigue. Biol Pharm Bull. 2006 May! 29 (5): 962-6.
- 15. Takasu J, Uykimpang R, Sunga MA, Amagare H, Niihara Y. Aged ALLIUM SATIVUM extract is a potential therapy for sickle-cell anemia. J Nutr. 2006; 136 (3 suppl): 803s-805s.