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Sulfur: A Natural Alternative To Synthetic Fungicides- A Review.

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Abstract:
The review addresses the efficacy of sulfur nanoparticles in antifungal treatments and examines the use of sulfur in traditional remedies, including its applications in dermatology. Special attention is given to sulfur compounds derived from *Allium sativum* (garlic), such as ajoenes, diallyl disulfide (DADs), vinyl dithiins, and S-allyl cysteine. These compounds are known for their antimicrobial and antifungal properties. Ajoenes, in particular, demonstrate strong inhibitory effects against fungal pathogens and exhibit potential for broader therapeutic applications. The review highlights the significance of sulfur in modern medicine and its potential as a natural alternative to synthetic fungicides.

Keywords: Antifungal efficacy, Sulfur, candida spp, Aspergillus fumigatus, Tropical preparation.

Introduction: There are two types of sulfur, organic sulfur and elemental sulfur. Due to its potent antimicrobial qualities, sulfur, a readily available physiologically active element, has been used for dermatological treatment since ancient times [1].

Sulfur is an abundantly available biologically active element, and has long been used for dermatological treatment since ancient times [2]. It's a natural component of induced in defence mechanism of antifungal, there are many S-containing compounds which have been linked directly or indirectly with the defence of plant against pathogens. Pathogens include thionine, defensins, glucosinolates, alliins [3].

It is noted that, at the basal level, sulfur plays a critical function in the machinery of protein synthesis. The bacteria are harmed by high sulfur concentrations. [4].

It frequently passes through an asexual stage in nature. Conidia are a type of airborne, uninucleate spore produced by A. fumigatus. The term "*Aspergillus"* refers to a group of respiratory illnesses that can be roughly divided into three main categories: invasive, allergic, and chronic. A. fumigatus is a source of novel targets for antifungals [5]. The involvement of glucosinolates in drug resistance and other sulfur-containing molecules, such as cysteine-rich antifungal protein, are both influenced by elemental sulfur antimicrobial action is shown via protein extracts [6]. In veterinary and medical settings alike, a high sulfur content is necessary for optimal treatment. Using sodium sulphite and *Catharanthus roseous* extract, sulfur nanoparticles were created at different pH levels [7]. Sporadic azole antifungal medication treatment is used to eradicate localized fungal infections on the surface of the epidermis and hair [8].

The fungus *Microsporum* cannis is the most frequent cause of dermatophytosis in animals [9]. It has been suggested that sulfur nanoparticles with antifungal action against Candida albicans could be synthesized using extract from *Allium sativum* [10].

In *Gandhak* rasayana, the percentage of sulfur has decreased from 98.26% to 5.61%. This decrease is associated with metabolic processes and detoxification [11]. Another method of treating mycosis and candidiasis is the widespread use of plant extracts. It can be found in *young ramson leaves*. At room temperature, allicin was unstable. The spectrum activity determines how effective it is [12]. certain sulfur-containing substances present in cabbage and the fermentation result. At Ph 5, 6, and 7, methyl methane thiosulfate exhibited 10 to 100 times greater inhibitory power against listeria monocytogens [13].

Targeting cysteine biosynthesis could also be a valuable strategy to fight against *aspergillus fumigatus.* A. fumigatus is a good target for antifungal drug development [14]. Sulphur uptake is required for incorporation into the sidechains of the amino acids cysteine and methionine, and is also essential for the biosynthesis of the antioxidant glutathione (GSH), S-adenosylmethionine (SAM) [15]. Anti-microbial of different organosulfur of products, such as thiosulfinates and thiosulfonates obtained from garlic (*Allium sativum*) and (*Allium cepa*) onion [16]. Sulfur is one of the substances whose biocide properties (including antifungal and anti-bacterial) have been indeed recognised for a long time. Wilkinson ointment and other are used for treatment of various skin diseases of parasitic, mycotic and several others [17]. The halogenomethylsulfonyl groups in it, which contain bromine and chlorine, are quite effective against C. albicians. The halogenomethylsulfonyl groups in it, which contain bromine and chlorine, are quite effective against C. albicians [18]. Dermatophytosis, sometimes known as ringworm, is a superficial skin disease. The purpose of sulfur is to lyse the fungi on the cell wall structure, destroying the fungus and causing fungal cell death [19]. Eighty percent of the amount of sulfur mustard evaporates upon contact with the skin, while the remaining twenty percent is absorbed and rapidly enters the keratinocytes and cell membranes of hair follicles. Of the absorbed quantity, 8% goes into the systemic blood circulation and 12% stays in the epidermis [20]. Many copper formulations have been used to treat skin problems, such as eczema, scars, syphilis, etc. Copper ions have previously been used to treat fungal infections [21].

2. Methodology: *Amalasara Gandhak* is thought to be appropriate for medical use and is available as a yellow crystal [22]. *Gandhak* and the majority of the other rasa elements are imported mostly from India because they are not available in Sri Lanka [23]. The *Rasa Jala Nidhi* states that the second stage should be completed in *dolayantra* [24]. Ghee is used in most of the sulphur *'shodhana'* procedure to avoid the blistering while melting and it also smeared on cloth to facilitate filtration without adhering to the cloth, it was not done in the first and final steps to obtain purified sulfur, the same procedure was applied to all samples [25].

Fig. 1. Preparation of *Eclipta alba* extract (a) cleaned plant (b) extract of *Eclipta alba* (c) vessel with extract [60].

Fig. 2. First step of *'Gandhak shodhana'* (a) crude sulphur (b) Melted sulphur powder (c) Melted sulphur poured into the prepared vessel [24].

Fig. 3. Second step of *'Gandhak shodhana'* (a) preparation of the vessels with sulphur obtained after first step (b) sulphur placed in steamer for 3 h (c) steamed sulphur [59].

3. Laboratory sulphur purification

The traditional laboratory purification process used recrystallization of the crude sulfur. 400 ml of xylene was heated with 40 g of crude sulfur powder while stirring continuously. The mixture was then filtered and allowed to cool slowly to room temperature to promote crystal formation. [25].

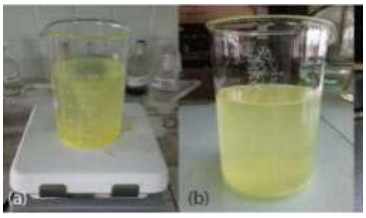

Fig.4 Sulphur laboratory purification (a) crude sulphur dissolved in xylene (b) filtered solution [57]

(c) recrystallization of sulphur (d) dried purified sulphur [57].

We used a hexaiodoplantinate reagent to modify a standard post-column HPLC technique. This technique is developed for the analysis of sulfur components in *aged* garlic extract (AGE) in a quantitative as well as qualitative manner [26].

A simple vacuum distillation process that makes it easier to analyse *Allium* volatiles qualitatively is explained [27]. According to recorded history, *Allium* sativum has been used for medical purposes for thousands of years, and Chinese medicine has been using it for at least 3,000 years [28].

4. Ajoene is a compound found in garlic and has several biological activities, including:

4.1 Anti-fungal activity: Ajoene has observed to have extremely strong inhibitory effects on the growth of *Aspergillus Niger* and *Candida albicans*. By altering the integrity of the cytoplasmic membrane, *ajoene* also inhibited the growth of the human fungal infection Paracoccidioides brasiliences [35].

4.2 Antiviral activity: When evaluated in an in vitro culture, the chemical ajoene reduced the tendency of the human immunodeficiency virus (HIV)-1 to respond to stimuli [36]. Compared to dextran sulphate, ajoene demonstrated a slightly higher level of activity in preventing HIV-1 virus cells from adhering to cells [37].

4.3 Anti-bacterial activity: Bacteria generate biofilms as part of their survival strategies. Bacteria grouped together in a self-produced matrix and attached to a surface or each other are called bacteria biofilms [38]. Proteins, polysaccharides, and extracellular DNA (eDNA) make up the matrix of biofilms. Therefore, biofilms serve as a barrier system that prevents bacteria from entering their cells to be affected by antibiotics, disinfectants, and host immune molecules, thereby resulting in drug resistance [39].

4.4 Antimicrobial actions of allicin: It has been discovered that allicin is active against *Staphylococcus aureus,* a human pathogen that is also resistant to methicillin, and that it efficiently inhibits the organism. *Allicin's* capacity to prevent the growth of specific microbes was investigated [40]. Another study that looked into how allicin works as an antibacterial discovered that it reacts with cysteine. The interaction led to the suppression of the organism's growth through a disulfide exchange-like reaction with the sulfhydryl-group of cysteine [41].

4.5 Anti-fungal activity: In addition to its antibacterial qualities, *allicin* is toxic to fungal cells and can prevent hyphal development and spore germination in both in vitro and in vivo cultures [42]. In an in vitro experiment employing fresh juice of *Allium sativum* and spore-seeded agar on a plate - diffusion assay - allicin has demonstrated strong inhibitory activities against plant-pathogenic fungal species such as *Plectospherella cucumerina, Botrytis cinerea, Magnaporthe grisea, and Alternaria brassicicola* [43]. Garlic juice treatment was seen to disinfect carrot seeds infected with *Alternaria brassicicola* in a way similar to that of conventional seed disinfection agents [44]. Apart from its application in agriculture*, allicin* has been linked to fungal infections in humans and animals. One theory proposes that allicin could form the basis for a treatment for lung aspergillosis infections due to its high volatility and potential for inhalation. [45]. It is also feasible to apply allicin topically to treat fungal skin infections. Allicin's efficacy was shown to be comparable to that of fluconazole, an antimycotic drug, in an attempt to treat a Candida infection [46].

4.6 Anti-hyperlipidaemic activity of allicin: Allicin treatment partially reversed the effects on rats whose low-density lipoprotein and low-density cholesterol levels had increased following an atherogenic diet [47]. The effects of garlic powder (0.6%) allicin on serum cholesterol and triglyceride levels revealed a significant drop [48].

5. Sulphur compound present in allium sativum.

5.1 Ajoenes (an unsaturated organosulfur compound found in small quantities)

Ajoenes are strong antibacterial agents that stop the growth of both bacteria and fungus. They have functional groups called sulfoxide and disulfide These substances have also been connected to *Allium satium*'s anticoagulant characteristics [30].

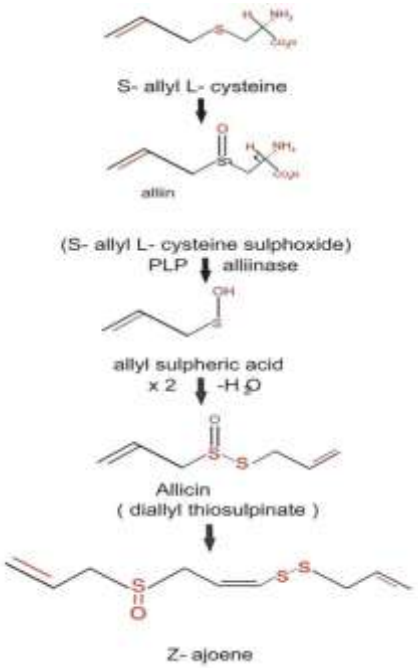

Fig.5 The biosynthesis of ajoene (z- isomer) [57].

The purpose of the screening was to find these naturally occurring QS inhibitors. Researchers found that *Allium sativum* toluene extract could prevent bacteria from forming biofilms. This suggests that *Allium sativum* extracts could have QS-inhibiting properties [31].
Ajoene also down-regulated the QS-regulated virulence factor genes involved in biofilm formation, namely chitinase, *Lasa* protease, lectin, endoprotease, and AB operon [32]. *Ajoenes* have been reported to be efficient against fungi, including *Aspergillus Niger* and *Candida albicans,* with the Z-isomer having slightly higher (2x) activity than the E-isomer [33]. A medication called *ajoenes* is presently being developed to treat blood coagulation issues [34].

5.2 Dially disulphide (DADs): Dially disulphides (DADs) are the main components *of Allium sativum* oil; they are created from allicin by steam distillation during the processing of garlic oil [49]. DADs have minimal germicidal activity against Gram-positive bacteria, and even drug-resistant bacteria [50]. The amount of sulphur atoms in a DAD molecule determines its antimicrobic activity; therefore, DADs with more than five atoms typically have more potent antibacterial activity [51].

5.3 Vinyl dithiins: The compounds known as vinyl-dithiins, also known as 3-vinyl-4H-1,2-dithiin and 2-vinyl-4H-1,3-dithiin, are unique sulfur-containing lipophilic compounds that are produced by the breakdown of allicin in macerate *Allium sativum* oil products [52]. These compounds have a number of biological activities, including antioxidant and anti-obesity properties, and there is evidence that they may also lower cholesterol levels [53].

5.4 S-allyl cysteine: S-allyl cysteine (SAC) is an organosulfur molecule that is hydrophilic and has a high bioavailability of nearly 100%. It is also safe and effective [54].
It is also observed that black *Allium sativum* has the same high content. SAC has been shown to be advantageous in aging and many disease models; these advantages are associated with its antioxidant processes [55].

Table1.1 Chemical properties of the sulphur containing compounds in allium sativum

Sr.no	Name	Chemical formulae	Molar_mass (m/mol)	features	Content in allium sativum (g)	stability
1.	Ajoene	$C_9H_{14}OS_3$	234.39	Colourless liquid	0.5-0.1	Very stable
2.	Allicin	$C_6H_{10}OS_2$	162.26	Yellowish liquid	2.5-4.5	Very unstable
3	DADs	$C_6H_{10}S_2$	146.28	Yellowish liquid	Not quantified	Unstable
4.	vinyl dithiins	$C_6H_6S_2$	142.24	Yellowish liquid	3.0	-
5.	S-allyl cystein	$C_6H_{11}NO_2S$	161.22	Crystalline solid	Not quantified	Highly stable

Conclusion: This is a review article discussing the use of sulfur as a natural alternative to synthetic fungicides. The antimicrobial properties of sulfur and its long history of use in dermatological treatments. It also explores the different forms of sulfur, including organic sulfur and elemental sulfur, and their role in the sulfur cycle. The mode of action of elemental sulfur and its effects on bacteria and fungi. It also examines the use of sulfur nanoparticles and their potential applications in antifungal treatments. And the use of sulfur compounds found in Allium sativum (garlic) for their antimicrobial and antifungal properties. These compounds include ajoenes, diallyl disulfide, vinyl dithiins, and S-allyl cysteine.

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