

The Bactericidal Effect of Aloe on Pathogens of Pneumonia

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Abstract. The respiratory tract encounters 10,000 liters of inhaled air every day. Recently, data have emerged on innate lung immunity and key pathways relevant to biomarker detection and therapeutic targeting strategies in acute and chronic respiratory diseases. The lungs serve as the main link between the host and the external environment. Thus, multiple lines of defense protect the host from inhaled potential pathogens. Impaired innate lung immunity can lead to detrimental outcomes such as pneumonia and disseminated infection. Various streptococci are of great ecological importance as part of the normal microbial flora of animals and humans; some of them can also cause diseases that range from subacute to acute or even chronic. Significant human diseases caused by streptococci include scarlet fever, rheumatic heart disease, glomerulonephritis, and pneumococcal pneumonia. Streptococci are essential in industrial and dairy processes and as indicators of contamination. The greatest attention is paid mainly to the species that cause severe infections: *S. pyogenes* and *S. pneumoniae* (pneumococci).

1 Introduction

The respiratory tract encounters 10,000 liters of inhaled air every day. While much of the air contains harmless environmental components, the lung's immune system must also cope with harmful microbial or sterile threats and respond quickly to protect the host in this barrier zone. The respiratory tract is endowed with a wide arsenal of cellular and humoral host defense mechanisms, most of which are related to the innate immune system. The complex interplay between resident and infiltrating immune cells and secreted innate immune proteins shapes the host-pathogen, host-allergen, and host-particle interactions in the respiratory mucosa. Recently, data have emerged on innate lung immunity and key pathways relevant to biomarker detection and therapeutic targeting strategies in acute and chronic respiratory diseases. The lungs serve as the main link between the host and the external environment. Thus, multiple lines of defense protect the host from inhaled potential pathogens. Impaired innate lung immunity can lead to detrimental outcomes such as pneumonia and disseminated infection. Innate lung immunity, the first line of defense, is provided by airway and alveolar epithelial cells, as well as by resident and recruited leukocytes. However, children under 3 years of age and older people over 65 years of age are constantly at risk, especially in winter. The lungs serve as the main link between the

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host and the external environment. Thus, multiple lines of defense protect the host from inhaled potential pathogens. Impaired innate lung immunity can lead to detrimental outcomes such as pneumonia and disseminated infection. The lungs are ventilated with thousands of liters of air per day.

The respiratory system inevitably comes into contact with airborne microbial compounds, most of which are harmless pollutants. It is known that epithelial cells of the respiratory tract have innate sensory functions that allow the detection of microbial hazards. To avoid chronic inflammation, the lung system has developed special tools to control local immune responses. Although airway epithelial cells may act as anti-inflammatory stimulants, it is hypothesized that, under homeostatic conditions, airway epithelial cells are important modulators of immune responses in the lung. The genus *Streptococcus*, a heterogeneous group of Gram-positive bacteria, is of wide medical and industrial importance. Various streptococci are of great ecological importance as part of the normal microbial flora of animals and humans; some of them can also cause diseases that range from subacute to acute or even chronic. Significant human diseases caused by streptococci include scarlet fever, rheumatic heart disease, glomerulonephritis, and pneumococcal pneumonia. Streptococci are essential in industrial and dairy processes and as indicators of contamination. The greatest attention is paid mainly to the species that cause severe infections: *S. pyogenes* and *S. pneumoniae* (pneumococci). Approximately 5-15% of people carry *S. pyogenes* or *S. agalactiae* in the nasopharynx. Pneumonia only infects humans, and no reservoir has been found in nature. All ages, races and genders are susceptible to streptococcal infections. Generally, pneumococcal infection is most prevalent in winter, coinciding with an increase in infection but not necessarily spread. The incidence of respiratory illness associated with *S. pyogenes* peaks around age 6 and then again at age 13, and is most common in late winter and early spring in temperate climates.

2 Research Methodology

Pneumococcal pneumonia is a life-threatening disease often characterized by swelling and rapid consolidation of the lobes. Recently, herbal remedies have been actively used to treat many bacterial infections, including pneumonia. Since the work is related to the effect of aloe juice on the bacterial cells of the pathogens of pneumonia, we will first dwell in more detail on the features of aloe. The Aloe genus is represented by 600 species and belongs to the Asphodelaceae family [1, 3]. Aloe plants are native to sub-Saharan Africa, on many islands in the western Indian Ocean, including Madagascar and the Saudi Arabian Peninsula. Aloe vera (AB) is a cactus-like plant that has been widely used in herbal medicines for thousands of years [2,5]. Compounds in the fleshy leaves, such as aloe emodin, aloin, aloesin, saponins, terpenoids, and polysaccharides, have wound healing, anti-inflammatory, immune, antidiabetic, antioxidant, laxative, antibacterial, antifungal, antiviral, and antitumor effects [1,4]. At present, AV is also widely used as cosmetic moisturizers, toothpastes, food flavors and preservatives in the pharmaceutical and food fields [2].

There are a lot of antibacterial agents, people actively use them, but over time they get used to them, and microorganisms are constantly rebuilding, adapting and eventually becoming resistant to them. The basis of many medicines includes various plant components. Recently, herbal remedies have been actively used to treat many bacterial infections, including pneumonia. The latex of the leaves of some aloe species and their constituents have a wide range of biological properties, such as antimicrobial [1,6], antimalarial [5,6] and antiglycation. The use of aloe vera is promoted in a huge number of diseases [5]. Aloe vera, which is a very common houseplant, is called the "house doctor" and is used in medicine throughout the globe [2,5]. The word vera means "real", "true" or

"authentic" [1]. Aloe vera is a widely used medicinal plant known for its diverse therapeutic applications [3] and has been used for several millennia in the medicine of some peoples. Aloe vera gel has anti-inflammatory as well as immunostimulating and immunomodulatory properties. Compounds in the fleshy leaves, such as aloe emodin, aloin, aloesin, saponins, terpenoids, and polysaccharides, have wound healing, anti-inflammatory, immune, antidiabetic, antioxidant, laxative, antibacterial, antifungal, antiviral, and antitumor effects [4,7]. Therefore, it seemed interesting to study the effect of aloe extracts on bacterial cells of aloe pathogens. The relevance of the problem lies in the study of the bactericidal characteristics of aloe on bacterial cells of pathogens of pneumonia, since this plant is medicinal and has long been practiced in natural treatment and serves as an alternative therapy for inflammatory diseases of the respiratory tract.

3 Results and Discussions

Aloe vera is one of the pharmaceutical herbs belonging to the lily family. It has been used in the treatment of various ailments, including dermatological infections, and has been used as a laxative since ancient times. This plant has long fleshy thick leaves with twisted sides that end in thorns [3]. Plants have a wide range of phytochemical composition. Some of the proven pharmacological actions that support the traditional use of each aloe species are due to the presence of a wide range of phytochemicals. The leaves of the aloe plant, which are the most commonly used medicinal parts, are heterogeneous and can be divided into three main parts, namely: the outer green epidermis, mainly composed of structural components; the outer region of the pulp under the epidermis, consisting of vascular bundles from which comes the bitter latex or juice; the pulp of the inner leaf, consisting of aloe gel and containing parenchyma cells. Regarding the different composition of these leaf parts, they probably also contain different classes of biologically active compounds that are thought to contribute to different leaf biological properties [1]. Briefly, the outer green epidermis is reported to contain anthraquinones, preanthraquinones, and their respective glycosides [2,7,6], while the outer pulp area below the epidermis contains a latex that is predominantly composed of phenolic compounds, including anthraquinones and preanthraquinones, anthrones, chromones, coumarins, flavonoids and pyrones [5,7]. On the other hand, the pulp of the inner leaf contains a high content of acemannan polysaccharide, as well as a wide range of phytochemicals, including alkaloids, anthraquinones, anthrones, chromones, coumarins, flavonoids, and pyrones [8,6,4]. The pulp also contains vitamins, minerals, enzymes and proteins [3,4]. Indeed, many authors believe that the different biological activities associated with different types of aloe should be attributed to the synergistic action of several compounds, rather than a single chemical [6,4,2].

The antimicrobial activity of *A. vera* juice was tested by Alemda and Agaoglu [5]. Dharajiyi et al. [8], the antibacterial activity of *A. barbadensis* leaf extract was evaluated against *E. coli*, *P. aeruginosa*, *B. cereus*, and *Serratia marcescens*. They found maximum inhibitory activity against *S. marcescens* (hexane extract) and *B. cereus* (methanol extract). In general, the authors noted that the methanol extract showed an inhibitory effect against all bacterial strains tested, while the ethyl acetate extract showed no inhibitory activity. Similar results of antibacterial activity against *E. coli*, *B. subtilis*, *S. epidermidis* and *S. sonnei* were obtained by Cooposamy and Magwa [2,3,8]. They found that the MIC of emodin and aloin A ranged from 62.5 mg/ml against *B. subtilis* and *E. coli* to 250 mg/ml against *S. epidermidis* and *S. sonnei* [2,7,6]. Juice and leaf extracts of *A.vera* were tested for antimicrobial properties by Abakar et al. [1].

To conduct studies to identify the effect of aloe juice on bacterial cells of pathogens of pneumonia, 15 strains obtained from sputum from patients with pneumonia were used. For the cultivation of pneumococci, both conventional culture media and media supplemented

with blood or serum are suitable. Growth is much faster on the latter media, as they prefer media supplemented with 0.1% glucose or 5% blood.

The juice of the Aloe vera plant, considered one of the most medicinal species, was the object of our research. Since the use of aloe aged from 3-5 years is desirable, it was these plants that were used. At this age, the concentration of nutrients and medicinal substances is considered the highest in aloe. Fresh leaves were used and had not been watered for seven days prior to pruning for study. For medicinal purposes, the lower or middle leaves, the largest and most fleshy, are usually used to obtain more juice. These are the leaves we used. Thoroughly washing the leaves, the peel was removed, carefully separating the gel. The aloe juice thus obtained in the amount of 5 grams was used for further research.

We used a universal nutrient medium, that is, meat-peptone agar, inoculated the obtained strains into Petri dishes, strictly observing all the rules. For incubation, they were placed in a thermostat at 37°C for a day. 1 ml of gel was added to the test sample, and the same medium, but without gel, was used as a control. The sensitivity of bacteria to aloe gel was determined as follows. Working dilutions of aloe juice were prepared, which were applied dropwise into Petri dishes, pre-distributed into sectors.

The conducted studies gave the following results. Tender, translucent colonies were obtained on dense nutrient media. The structure of the colonies is very clear, as if outlined, they do not merge with each other, the diameter of the colonies is about 1 mm.

Streptococcus pneumoniae often forms short chains of ovoid diplococci, and α -type colonies on blood agar. Chains usually consist of 2-8 pairs of cocci. Pneumococci are surrounded by a polysaccharide capsule. They are rarely spherical, more often they resemble short artillery shells in shape, touching their bases. The causative agent of diplococcal infection has the form of paired cocci, the latter being somewhat elongated in length, their outer ends are lanceolate-pointed (they are also compared with the contours of a candle flame). In the pathological material (smears from affected tissues, sputum, blood), pneumococci have a clearly visible capsule covering both cocci. In cultures of diplococcus, the capsule is usually absent and often the microbe forms short chains, while acquiring similarities with streptococcus. The size of individual cocci ranges from 0.5 to 1.5 microns.

Pneumococci stain well with conventional aniline dyes, are gram-positive, immobile, do not form spores. The capsule with the usual color of pneumococcus remains colorless (they do not perceive paint); it is well stained according to Romanovsky - Giemsa.

Studies on the bactericidal activity of aloe using various concentrations from a minimum of 12.5 mg / ml to 100 mg / ml gave the following results. Quite high bactericidal activity of aloe extract was tested by strains numbered 1,2,3,4,8,10, 11,12,13 (Fig. 1).

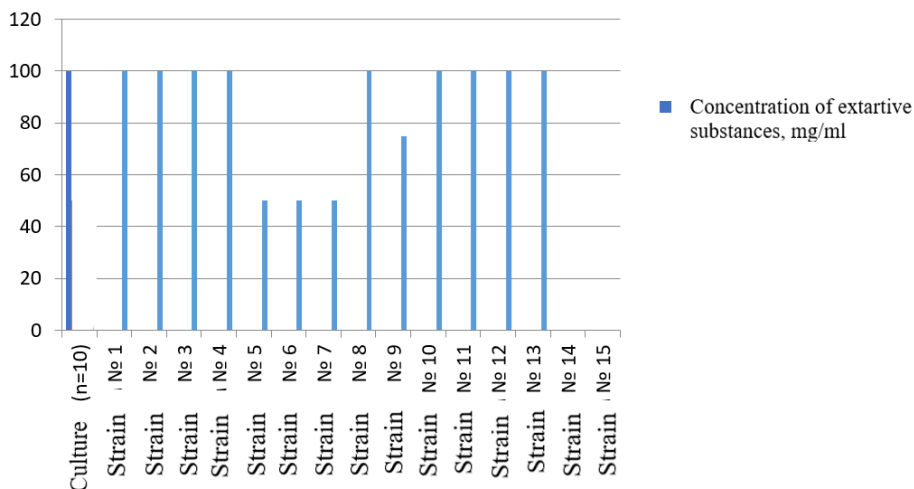


Fig. 1. Activity of pneumococci depending on the concentration of aloe

A weaker effect was on strain No. 9 (75%), strains No. 5,6,7 even weaker (50%), strains No. 14 and 15, respectively - 0%, that is, there was no bactericidal effect at all. Further results of the study of the bactericidal action of the aloe extract were more obvious, since the number of grown colonies was significantly reduced. This feature is strongly noticeable in comparison with the control sample. The results are summarized in table 2, which summarizes the data on the suppression of the growth of test cultures of pneumococci prepared with an extract of real aloe juice (MIC - minimum inhibitory concentration). This stage of research showed a pronounced microbicidal effect of real aloe juice on bacterial cells of pneumonia pathogens. Compared to the control sample, where a huge number of colonies grew, in Petri dishes with aloe extract, there are significantly fewer colonies.

Figure 2 shows the dependence of the number of colonies on the concentration of 50 mg/ml.

The conducted studies have convincingly proved that the extract of real aloe juice has pronounced bactericidal properties and inhibits the growth of pathogenic flora.

Streptococcus pneumoniae is a major human pathogen causing diseases such as pneumonia and meningitis. New methods and technologies in the field of lung biology are beginning to shed new light on the microbial world of the respiratory tract. Long considered a sterile environment, it is now clear that human lungs are often exposed to live microbes and their by-products. The nature of the microbiome of the lungs is very different from other microbial communities inhabiting our body, for example, in the intestines. Notably, the lung microbiome has low biomass and is dominated by dynamic flows of microbial immigration and clearance, resulting in bacterial load and microbiome composition that are inherently variable rather than fixed. The dynamic nature of the lower respiratory tract microbiome is constantly evolving. The main factors that control the balance of the microbiota are the immigration of microorganisms by inhalation and microaspiration, as well as the elimination of microorganisms by coughing, mucociliary transport and immune responses.

The lungs serve as the main link between the host and the external environment. Thus, multiple lines of defense protect the host from inhaled potential pathogens. Impaired innate lung immunity can lead to detrimental outcomes such as pneumonia and disseminated infection. The lungs are ventilated with thousands of liters of air per day. The respiratory system inevitably comes into contact with airborne microbial compounds, most of which are harmless pollutants. It is known that epithelial cells of the respiratory tract have innate

sensory functions that allow the detection of microbial hazards. To avoid chronic inflammation, the lung system has developed special tools to control local immune responses. Although airway epithelial cells may act as anti-inflammatory stimulants, it is hypothesized that, under homeostatic conditions, airway epithelial cells are important modulators of immune responses in the lung. The genus *Streptococcus*, a heterogeneous group of Gram-positive bacteria, is of wide medical and industrial importance. Various streptococci are of great ecological importance as part of the normal microbial flora of animals and humans; some of them can also cause diseases that range from subacute to acute or even chronic. Significant human diseases caused by streptococci include scarlet fever, rheumatic heart disease, glomerulonephritis, and pneumococcal pneumonia. Streptococci are essential in industrial and dairy processes and as indicators of contamination. The greatest attention is paid mainly to the species that cause severe infections: *S. pyogenes* and *S. pneumoniae* (pneumococci). Approximately 5-15% of people carry *S. pyogenes* or *S. agalactiae* in the nasopharynx. Pneumonia only infects humans, and no reservoir has been found in nature. All ages, races and genders are susceptible to streptococcal infections. Generally, pneumococcal infection is most prevalent in winter, coinciding with an increase in infection but not necessarily spread. The incidence of respiratory illness associated with *S. pyogenes* peaks around age 6 and then again at age 13, and is most common in late winter and early spring in temperate climates.

4 Conclusions

1. During the research, a pure culture of *Streptococcus pneumoniae* was obtained from the material provided by the bacteriological laboratory. Tender, translucent colonies were obtained on dense nutrient media. The structure of the colonies is very clear, as if outlined, they do not merge with each other, the diameter of the colonies is 1 mm. *Streptococcus pneumoniae* often forms short chains of ovoid diplococci, and α -type colonies on blood agar. Chains usually consist of 2-8 pairs of cocci.

2. We got the gel from the real aloe plant.

3. When studying the bactericidal activity of aloe juice using various concentrations from a minimum of 12.5 mg / ml to 100 mg / ml, the following results were obtained. Quite high bactericidal activity of aloe extract was tested by strains numbered 1,2,3,4,8,10, 11,12,13. A weaker effect was on strain No. 9 (75%), strains No. 5,6,7 even weaker (50%), strains No. 14 and 150%, that is, there was no bactericidal effect at all. Further results of the study of the bactericidal action of the aloe extract were more obvious, since the number of grown colonies was significantly reduced. This feature is strongly noticeable in comparison with the control sample.

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