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Research Article

EFFICACY OF ASTRAGALUS EXTRACT AGAINST *S. mutans* and *C.albicansan* *in vitro* Study

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In this *in vitro* Microbiologic study was done using Mitis Salivarius agar media (for *S.mutans*), Sabouroud Dextrose agar media (for *Candida albicans*)

ABSTRACT

Aim- To check the efficacy of Astragalus extract against *Streptococcus mutans* and *Candida albicans*

Methodology- In this *in vitro* Microbiologic study was done using Mitis Salivarius agar media (for *S.mutans*), Sabouroud Dextrose agar media (for *Candida albicans*) for selective growth of different microorganisms via Kirby Bauer disk diffusion method. Freshly prepared media was used and allowed to dry, over that microorganism was taken from the prepared strains of Streptococcus mutans and Candida albicans which was streaked uniformly over medias, after that paper discs which were dipped overnight in the prepared Astragalus extract solution, Chlorhexidine solution and distilled water were placed over media and labelled 1, 2 & 3 respectively. After 24 hours of incubation the medias were checked for zone of inhibition.

Result- Astragalus showed good anti-microbial activity against *S. mutans* and its activity was somewhat similar to that of chlorhexidine in relation to *S. mutans* and even more effective in some samples, but astragalus' activity against *C. albicans* is relatively less effective although it provides certain amount of anti-microbial action against it too

Conclusion- Astragalus inclusion in diet or in oral hygiene practices methods such as an ingredient in dentifrices or in mouthwashes can prove to be beneficiary. Its antimicrobial efficacy in addition to number of other contributory effects such as anti-inflammatory, anti-carcinogenic, immunomodulatory actions can enhance the host response.

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INTRODUCTION

Astragalus comes from a large genus of herbs which consists of about 3,000 species of herbs and small shrubs. It belongs to the legume family Fabaceae and subfamily Faboideae.¹ A native to Northern Hemisphere, it is commonly known as Milkvetch (most species), locoweed, and Goat's thorn and in China commonly known as Huang Qi. Other names for astragalus are green dragon, gum dragon, gummitragacanth, hog gum, membranous milk vetch.²

It is a sweet herb with its particular effect on lung, spleen and heart. It is basically used in the treatment of fatigue, loss of appetite, viral infections, fever, etc. But it has been majorly used as an immunomodulator to fight against viral illness; it has been a subject of research and has been used for its action against cancer, HIV and atopic diseases. Astragalus has been subjected to be used as an additive to other herbs instead of a

single agent. It is often combined with ginseng, angelica, liquorice and other herbs.³

There are numerous species of Astragalus genus. The main species which is used medicinally is *Astragalus membranaceus*, whereas *Astragalus trigonus* and *Astragalus gummifera* have also been used.⁴

Astragalus is also known as an 'adaptogen' as it helps protect body against various stresses including physical, mental, or emotional stress.

Astragalus has number of effects on human health such as acting as an anti-inflammatory, boosting immune system, anti-aging effect, protects the cardiovascular system, helps fight against diseases such as cancer, HIV, etc.^{5,6,7,8}

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METHODOLOGY

Study conducted at: Department of Microbiology, Rungta College of Dental Sciences & Research, Kohka-Kurud Road, Kohka, Bhillai.

Study Design: in-vitro

An *in vitro* Microbiologic study was conducted to test the efficacy of Astragalus root extract against *S. mutans* and *C. albicans* with the help of Mitis Salivarius agar media (for *S. mutans*) and Sabouroud Dextrose agar media (for *C. albicans*) for selective growth of the previously mentioned microorganisms by using the Kirby Bauer disk diffusion method. The extract to be tested is prepared by mixing the Astragalus extract (®Ayurish- Astragalus extract) with distilled water in 1:1 proportion that is 1 gram of extract with 1 millilitre of distilled water, the positive control taken was Chlorhexidine 0.2 % (®Rexidin), and the negative control being distilled water. The prepared extract, positive control and negative control discs were prepared by immersing paper discs in the solutions and keeping it for a day to enable it to soak the solution then taking the discs out and allowing it to dry while keeping it in a separate container overnight.

Freshly prepared media of Mitis Salivarius agar media (HiMedia) and Sabouroud Dextrose agar media (HiMedia) were prepared using distilled water in a conical flask, the prepare media were then sterilized in an autoclave at a temperature of 121° C for 15 minutes under 15 psi pressure to rule out any contamination, it was then taken out and poured into petri dishes inside the laminar air flow chamber and were allowed to dry so as they solidifies for further procedure (for each microorganism separate 10 petridishes were prepared that is each *S. mutans* and *C. albicans* consisting of 10 samples), over that microorganisms were taken from the prepared strains of *S. mutans* and *C. albicans* which were streaked uniformly over medias, and after that paper discs which were dipped overnight in the different preparations of Astragalus extract solution, Distilled water (as a negative control) and Chlorhexidine solution (as a positive control) were placed over media and labelled 1, 2 & 3 respectively. After 24 hours of incubation time the media were checked for zone of inhibition using HiMedia zone reader scale.



Figure 1 Showing (Left) Astragalus extract and its prepared discs, (Centre) Distilled water and its paper discs and (Right) Chlorhexidine 0.2% and its prepared discs.



Figure 2 Showing (Left) Strain of *C. albicans* and (Right) Strain *S. mutans*

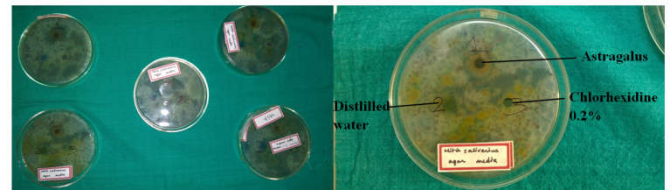


Figure 3 Showing zone of inhibition created by prepared extract and controls against *S. mutans*; (1- Astragalus extract), (2- Distilled water) and (3- Chlorhexidine 0.2%)

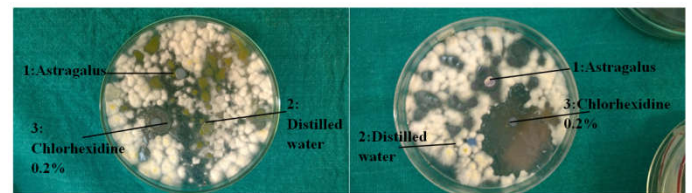


Figure 4 Showing zone of inhibition against *C. albicans*; (1- Astragalus), (2) Distilled water, (3- Chlorhexidine 0.2%)

RESULT

The Astragalus showed good anti-microbial activity against *S. mutans* and its activity was somewhat similar to that of chlorhexidine in relation to *S. mutans* and even more effective in some samples, but astragalus' activity against *C. albicans* is relatively less effective although it provides certain amount of anti-microbial action against it too

Table 1 represents the Comparison of mean of Astragalus test in Streptococcus Mutans and Candida Albicans at 24 hours. It was evident from the 24 hour test that the mean (S.D.) in *S. mutans* was 12.8 ± 1.8 and in *C. albicans* group was 6.6 ± 0.84 ($P = 0.001$).

Table 2 represents the Comparison of mean of Chlorhexidine test in Streptococcus Mutans and Candida Albicans at 24 hours. It was evident from the 24 hour test that the mean (S.D.) in *S. mutans* was 12.6 ± 1.4 and in *C. albicans* group was 9.6 ± 1.5 ($P = 0.001$).

Table 3 represents Comparison of mean of Distilled water test in Streptococcus Mutans and Candida Albicans at 24 hours. It was evident from the 24 hour test that the mean (S.D.) in *S. mutans* was 0.4 ± 0.8 and in *C. albicans* group was 0.4 ± 0.5 .

Table 1 Comparison of mean of Astragalus test in Streptococcus Mutans and Candida Albicans at 24 hours

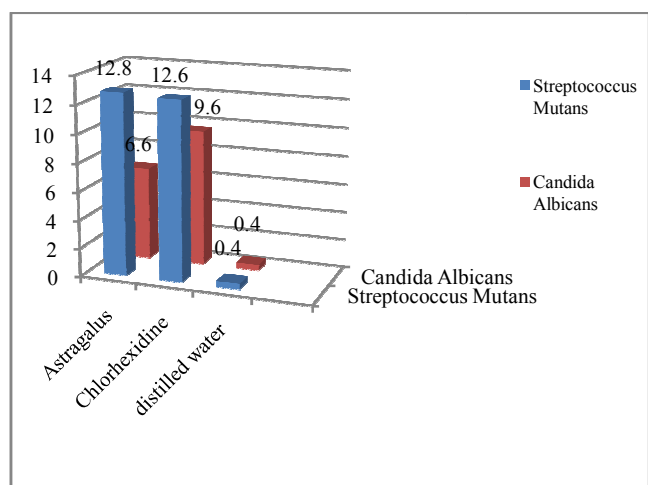
Test among group	N	Mean	Standard deviation	t- value	p- value
Streptococcus Mutans	10	12.8000	1.81353	9.803	0.001
Candida Albicans	10	6.6000	0.84327		

Table 2 Comparison of mean of Chlorhexidine test in Streptococcus Mutans and Candida Albicans at 24 hours

Test among group	N	Mean	Standard deviation	t- value	p- value
Streptococcus Mutans	10	12.6000	1.42984	4.456	0.001
Candida Albicans	10	9.6000	1.57762		

Table 3 Comparison of mean of Distilled water test in Streptococcus Mutans and Candida Albicans at 24 hours.

Test among group	N	Mean	Standard deviation	t- value	p- value
Streptococcus Mutans	10	.4000	.84327	1.0	0.001
Candida Albicans	10	.4000	.51640		



Graph 1 Showing comparative evaluation of Astragalus with Chlorhexidine and Distilled water against S. mutans and C. Albicans

DISCUSSION

Extracts of Astragalus are basically used as a life prolonging extracts for human use. Astragalus has a significant age reversal effect on our immune system; it declines the percentage of cytotoxic T cells and natural killer cells after 6 months to a year of use. Because of its anti-inflammatory qualities, astragalus has been in use for treating wounds.⁹ Radix astragali that is the dried root of astragalus, used in Traditional Chinese Medicine for the repair and regeneration of injured organs and tissues. A 2012 study conducted by the Institute of Pharmaceutics at Zhejiang University showed that wounds treated with astragaloside IV (the active ingredient in dried astragalus root) showed recovery rates increase two- to three times over 48–96 hours. It was concluded that astragalus is a promising natural product for anti-scarring and healing in wounds.

¹⁰Astragalus contains three main components that have such an effect on human health, these are as follows¹¹:-

1. Saponins
2. Flavonoids
3. Polysaccharides

Saponins are known for their ability to lower cholesterol, improve the immune system and prevent cancer.¹²

Flavonoids, also found in astragalus, provide health benefits through cell signaling. They show antioxidative qualities, control and scavenge of free radicals, and can help prevent heart disease, cancer and immunodeficiency viruses.¹³

Polysaccharides are known to have antimicrobial, antiviral and anti-inflammatory capabilities, among other health benefits.¹⁴

As of now we are familiar with the effects of astragalus its anti-bacterial and anti-fungal effect, it is due to the contribution of the above active components.

Saponins present in Astragalus contain something called astriterpenoidglycone with one or more sugar chains are attached to it. They are nonionic detergents which belong to the group of glycosides. Chemical structures determine their biological properties as natural nonionic detergents which have cytotoxic, hemolytic, molluscicidal, anti-inflammatory, antifungal, antiyeast, antibacterial, and antiviral activities. In a study, it has been suggested that saponin might disturb the permeability of the bacterial outer membrane. Saponin have surface-active properties, with capacity to penetrate into the lipid bilayer which further binds to cholesterol forming cholesterol-saponin complexes which finally lyses cells. About 90% of the surface of naturally cholesterol-free Gram-negative bacteria cell-wall outer membranes are covered by lipopolysaccharide (LPS). It then established that saponin might interact with the lipid-A part of Proteus LPSs and thereby increasing the permeability of bacterial cell wall. Lipid A-saponin complexes might promote antibiotic (colistin, ampicillin) uptake to inherently resistant bacteria cells.^{15, 16, 17, 18}

Flavonoid which is one of the active components of astragalus is a potent antioxidant, anti-inflammatory, anti-allergic, anti-carcinogenic, anti-hypertensive and anti-thrombotic agent. Several studies prove that these actions of flavonoid are attributable to its antioxidant property which traps the ROS (reactive oxygen species), chelates metallic ions and also contributes to the inhibition of enzymes responsible: xanthine oxydase.^{19, 20, 21}

Astragalus polysaccharides (APSs) represents itself as one of the most important natural active materials harvested out of astragalus and are very effective in traditional medicine due to its multitargeting biological activities which includes properties such as antioxidant, anti-inflammatory, anti-hepatitis B virus (HBV) and immune regulation effects.²²

Combination of all these three active components which are evidently anti-microbial in nature can prove to be of great significant, Astragalus thus possessing such characteristics may prove to be beneficial amongst other contemporaries as it provides immense versatility in order to its actions.

CONCLUSION

Herbal extracts provide an efficient way to fight against microorganism with no or minimal side effects to the human body, astragalus being one of the commonly used product of Traditional Chinese medicine have been widely used as an immunomodulator but this study shows that it can also be used as an effective anti-microbial agent against microorganisms present in oral cavity. Inclusion of astragalus extract in diet or in varied forms such as toothpaste, ointment, mouthwash etc can be beneficial in prevention from diseases such as dental caries, candidiasis, etc. Its advantage of having so many qualities such as immunomodulation, anti-cancer, anti-aging can only contribute and help in preventing other diseases too like leukoplakia, OSMF and others. Further studies or research has to be done to see the extent of its usage as a medicine for different entities and how it responds against various pathogens of oral cavity.

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