



## RESEARCH ARTICLE

### ANTIBACTERIAL ACTIVITY OF APPLE CIDER VINEGAR AGAINST CLINICAL ISOLATES OF *STAPHYLOCOCCUS AUREUS*

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#### ARTICLE INFO

##### Article History:

Received 22<sup>nd</sup> February, 2017

Received in revised form

18<sup>th</sup> March, 2017

Accepted 26<sup>th</sup> April, 2017

Published online 19<sup>th</sup> May, 2017

#### ABSTRACT

The rising colonization rates of *S. aureus* lead to the increasing of infection rates in the community and in hospitals. *Staphylococcus aureus* is one of the most frequent bacterial pathogen in humans. It causes skin infections, osteoarthritis and respiratory tract infections in the community, as well as postoperative and catheter-related infections in hospitals. The aim of the study was to determine the antibacterial activity of apple cider vinegar against clinical isolates of *S. aureus*. It did not show any inhibitory activity against *S. aureus* isolates. May be increased dilutions are required to standardize the efficacy.

#### Key words:

*Staphylococcus aureus*, MIC,  
Apple Cider Vinegar.

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Citation: Shreya Svitlana A. and Dr. Gopinath, P. 2017. "Antibacterial activity of apple CIDER vinegar against clinical isolates of *Staphylococcus aureus*", *International Journal of Current Research*, 9, (05), 49979-49981.

## INTRODUCTION

The rising colonization rates of *S. aureus* lead to the increasing of infection rates in the community and in hospitals. (Niyas and Gopinath, 2016) *Staphylococcus aureus* is one of the most frequent bacterial pathogen in humans. It causes skin infections, osteoarthritis and respiratory tract infections in the community, as well as postoperative and catheter-related infections in hospitals. (Didier *et al.*, 2004) *Staphylococcus aureus* is a gram-positive bacterium which is responsible for increased morbidity and mortality. (De Albuquerque, 2006) Recent reports of strains of methicillin-resistant *Staphylococcus aureus* (MRSA) isolated from children in the community have led to speculation that the epidemiology of *S. aureus* is changing. (Herold *et al.*, 1998; CDC, 1999; Boyce, 1998) Humans are a natural reservoir for *S. aureus*, and asymptomatic colonization is far more common than infection. Colonization of the nasopharynx, perineum, or skin, particularly if the cutaneous barrier has been disrupted or damaged, may occur shortly after birth and may recur anytime thereafter. (Payne *et al.*, 1966) Transmission occurs by direct contact to a colonized carrier.

Carriage rates are 25% to 50%; higher rates than in the general population are observed in injection drug users, persons with insulin-dependent diabetes, patients with dermatologic conditions, patients with long-term indwelling intravascular catheters, and health-care workers. (Transmission occurs by direct contact to a colonized carrier). The lack or loss of resistance to multiple antibiotics suggests a community origin because antibiotic selective pressure is much lower within the community than in hospitals, and the survival advantage of multiple-drug resistance is lower. (Chambers, 2001) In recent studies, essential oils and extracts derived from natural plant species have been known to produce anti-microbial property against virulent genus of micro-organisms. Investigations concerning the evaluation of the biological activities of essential oils of some medicinal plants have revealed that some of them exhibited antibacterial, antifungal and insecticidal properties. (Burt, 2004) In this study, we tried to assess the anti-bacterial effect of Apple Cider Vinegar against *S. aureus*. Scientists have measured ninety different substances in apple cider vinegars such as thirteen types of carboxylic acids, four aldehydes, twenty ketones, eight types of alcohols, eight ethyl acetates etc. It also contains important minerals, trace elements and vitamins (as listed underneath) as well as acetic acid, propionic acid, lactic acid and malic acid, enzymes, amino acids as well as roughage in the form of potash and apple pectin. Minerals and trace elements: Potassium, Calcium, Magnesium, Phosphorous, Chlorine, Sodium, Sulfur, Copper, Iron, Silicon, Fluorine. Vitamins: Vitamin C, Vitamin E,

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Vitamin A, Vitamin B1, Vitamin B2, Vitamin B6, Provitamin beta-carotene. (Jabir *et al.*, 2011) Apart from its anti-bacterial property, it also possesses anti-fungal, anti-viral, anti-cancer properties. It has various benefits like anti-aging, eliminating bad cholesterol; Polyphenols such as chlorogenic acid which is present in high levels in apple cider vinegar could inhibit oxidation of LDLs and improve health by preventing cardiovascular diseases (Laranjinha *et al.*, 1994), and helps in weight reduction. Thus, the aim of the study was to determine the antibacterial activity of apple cider vinegar against clinical isolates of *S. aureus*.

## MATERIALS AND METHODS

### Bacterial isolates

A total of 20 clinical isolates of *S. aureus* were collected from different clinical specimens of patients attending Saveetha Medical Collage and hospital. They were processed for a battery of standard biochemical tests and confirmed. Isolates were preserved in semisolid trypticase soy medium and stored at 4°C until further use.

### Antibiotic susceptibility test

Antibiotic susceptibility testing was determined for these isolates to the following antibiotics such as penicillin, erythromycin, clindamycin, ciprofloxacin, tetracycline, cotrimoxazole and linezolid. These antibiotics were procured from Himedia, Mumbai. This was performed by Kirby-bauer disc diffusion method as per CLSI guidelines. (Clinical and Laboratory Standards Institute, 2015)

### Detection of antibacterial activity of apple cider vinegar against clinical isolates of *S. aureus*

Antibacterial activity of apple cider vinegar was tested against *S. aureus* isolates by minimum inhibitory concentration method. Agar dilution method was performed to attain the different concentrations of apple cider vinegar such as 0.03%, 0.06%, 0.125%, 0.25%, 0.5%, 1% and 2% in MHA. Media containing various concentrations of vinegar were poured over the sterile petridishes and allowed to dry. Media without vinegar was served as control plate. Spot inoculation of 0.5 McFarland standard turbidity adjusted isolates were made on the plates and incubated at 37°C for overnight. The lowest concentration of the agent that completely inhibited the growth of isolates was considered as MIC. (Gopinath Prakasam *et al.*, 2014)

## RESULTS

### Sample wise distribution of clinical isolates of *S. aureus*

Of 20 clinical isolates of *S. aureus*, 8/20 (40%) were obtained from pus, 6/20 (30%) were from wound, 4/20 (20%) and 2/20 (10%) were from blood and sputum respectively (Figure 1).

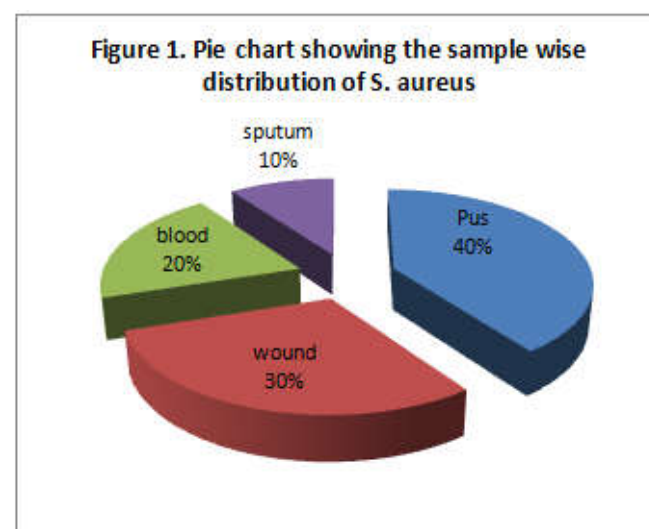
### Antibiotic susceptibility pattern

We have observed a varied pattern of sensitivity among one *S. aureus* isolates. There was complete resistance observed for penicillin (100%), 9/20(45%) isolates were shown to the resistant to erythromycin, 6/20(30%) were to cotrimoxazole, 4/20(20%) were to linezolid followed by 3/20(15%) were

resistant to ciprofloxacin and clindamycin respectively (Table 1).

**Table 1. Results of antibiotic susceptibility pattern of *S. aureus***

Antibiotics	Sensitive(%)	Intermediate(%)	Resistant(%)
Penicillin	0	0	20(100)
Erythromycin	14(70)	4(20)	2(10)
Clindamycin	15(75)	2(10)	3(15)
Ciprofloxacin	9(45)	8(40)	3(15)
Tetracyclin	14(70)	4(20)	2(10)
Cotrimoxazole	10(50)	4(20)	6(30)
Linezolid	10(50)	6(30)	4(20)



**Figure 2. Representative picture showing antibiotic sensitivity pattern of *S. aureus***

### Result of antibacterial activity of apple cider vinegar against clinical isolates of *S. aureus*:

It did not show any inhibitory activity against *S. aureus* isolates.

## DISCUSSION

Vinegar, alone, has been used for cleaning and treating nail fungus, head lice, warts and ear infections. Consumers typically prefer natural preservative methods for inhibiting the growth of food borne pathogenic microorganisms in food. (Finkel *et al.*, 2009) Several problems might be encountered with antifungal drugs, first; resistance; as fungi may become resistant to antifungal drugs due to target gene mutations, enzymemodification or to development of pump system that expels the drug out of the fungal cell (Higgins and Brinkhaus, 1999) second; toxicity; as antifungal drugs can cause systemic toxicities as hepatotoxicity and nephrotoxicity. These problems necessitate searching for safer effective remedies with known antifungal activity. (Rauha *et al.*, 2000)

Apple cider vinegar is a commonly prescribed antifungal agent in folkmedicine for treatment of fungal skin, ear and vaginal infections. (Rauha *et al.*, 2000) The antifungal activity of apple cider vinegar might be attributed to its malic acid, acetic acid contents or to other non-identified ingredients. The mechanism of inhibition fungi growth by organic acids is generally not considered a pH phenomenon. It is known that, growth and morphology of fungi are influenced by the pH of media. (Kang *et al.*, 2003) Organic acids resulting a decreasing in pH value,

this may influence the growth by acidifying the cell, which will consume a great amount of energy to maintain the intracellular pH homeostasis. (Hassan *et al.*, 2015) Other explanations have also been proposed including the membrane disruption, the interruption of metabolic reactions, and the accumulation of toxic anions. (Pelaez *et al.*, 2012) The inhibition of microbial growth increases by lowering pH of the media, and most microorganisms are susceptible to antimicrobial effects in the presence of organic acids. This phenomenon is due to the hydrophobic feature of most organic acids, which allows free diffusion of the protonized form through cell membrane. This diffusion process takes place spontaneously due to pH and osmolarity gradients that exist between the inner and outer sides of the cell. The intracellular pH is higher than the extracellular, and the acid undergoes dissociation as soon as it enters the cytoplasm and then decreases the intracellular pH by releasing the proton. To counter the decrease of cytoplasmic pH, resulting from the ionization of the entered acid, the cell allocates the main part of its energy content to eliminate these newly formed protons which results in slower growth kinetics. (Hassan *et al.*, 2015) According to Hassan *et al.* (Pelaez *et al.*, 2012), acetic acid shows strongest inhibition of fungal growth among other organic acids. Similarly, the study done by Prakasam *et al.* (Ravikumar *et al.*, 2016; Ravikumar, 2016) stated that natural products like aloe vera showed better inhibition when compared to calcium hydroxide. In this study,

## Conclusion

It did not show any inhibitory activity against *S. aureus* isolates. May be increased dilutions are required to standardize the efficacy.

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