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Investigation of *In vitro* Antimicrobial Activity of *Aloe vera* Juice

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Abstract: This study was conducted to determine the antimicrobial activity of the *Aloe vera* juice against Gram-positive bacteria (*Mycobacterium smegmatis*, *Staphylococcus aureus*, *Enterococcus faecalis*, *Micrococcus luteus* and *Bacillus sphericus*), Gram-negative bacteria (*Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, *Escherichia coli* and *Salmonella typhimurium*) and *Candida albicans* as *in vitro*. The disc diffusion method was used to test the antimicrobial activity. The study showed that *Aloe vera* juice has antimicrobial activity against *M. smegmatis, K. pneumoniae, E. faecalis, M. luteus, C. albicans* and *B. sphericus*, but has no inhibitory effect against the other bacterial strains. The least inhibitory effect was found against *M. luteus*, while *C. albicans* was detected to be the most sensitive strain. As a result; the antibacterial activity of the tested plant juice was found effective mainly against the Gram-positive bacteria. This is important to use of *Aloe vera* for medication, cosmetic and food purposes.

Key words: *Aloe vera* juice, antimicrobial activity, microorganism

INTRODUCTION

In many parts of the world, there is a rich tradition in the use of herbal medicine for the treatment of many infectious diseases (Brantner and Grein, 1994). In developing countries, it is estimated that about 80% of the population rely on traditional medicine for their primary health care (Matu and Van Staden, 2003). Because of the side effect and the resistance that pathogenic microorganisms build against the common antibiotics, much recent attention has been paid to extracts and biologically active compounds isolated from plants used in herbal medicine (Essawi and Srour, 2000).

Aloe barbadensis Miller (*Aloe vera*) belongs to the Liliaceal family, of which there are about 360 species. It is a cactus-like plant that grows readily in hot, dry climates and currently, because of demand, is cultivated in large quantities (Newall *et al*., 1996). Succulent, almost sessile perennial herb; has leaves 30-50 cm long and 10 cm broad at the base; colour pea-green (when young spotted with white); bright yellow tubular flowers 25-35 cm in length arranged in a slender loose spike; stamens frequently project beyond the perianth tube (WHO, 1999).

Aloe vera contains over 75 nutrients and 200 active compounds, including vitamins, enzymes, minerals, sugars, lignin, anthraquinones, saponins, salicylic acid and aminoacids (Park and Jo, 2006). Numerous scientific studies on *Aloe vera* are demonstrating its analgesic, anti-inflammatory, wound healing, immune modulating and anti-tumor activities as well as antiviral, antibacterial and antifungal properties (Anonymous, 2008).

Aloe vera products are used mainly for cosmetic, pharmaceutical and food industries (Eshun and He, 2004). Furthermore, these aloe species are currently listed in the pharmacopoeia of many countries in forms of plain aloe, extract and/or powder (Park and Jo, 2006). The industry size for aloe raw material is estimated to be about $125 million dollars. The volume of the industry for finished products containing *Aloe vera* is alleged to be around $110 billions dollars (Anonymous, 2006).

The aim of this study, was to determine whether *Aloe vera* juice has antimicrobial effect against microorganisms, including pathogens and spoilage-causing microorganisms.

MATERIALS AND METHODS

The juice from cold pressed leaves of *Aloe vera* plant (ALOVA-Australian import Traders GmbH.) was used as material. It was obtained from Doktor Klaus (IASC/Köln/Germany) and used without dilution in the trial of antimicrobial activity.

Test organisms: Five Gram-positive bacteria, 4 Gram-negative bacteria and one yeast culture were used to test strains. The bacterial strains of *Pseudomonas aeruginosa*

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(ATCC 25853), *Mycobacterium smegmatis* (CCM 2067), *Klebsiella pneumonia*e (FML 5), *Staphylococcus aureus* (ATCC 25923), *Enterococcus faecalis* (ATCC 15753) and *Micrococcus luteus* (A 2971) were obtained from culture collection of Department of Microbiology, Medical Faculty of Yuzuncu Yil University; *Escherichia coli* (ATCC 25922), *Salmonella typhimurium* (KUEN 1357) and *Candida albicans* (ATCC 60192) form Culture Collection of Industrial Microbiology and Biotechnology (KUKEM); and *Bacillus sphericus* (RSKK 384) from culture collection of Refik Saydam Health Institution of Health Ministry, Ankara, Turkey.

Antimicrobial susceptibility testing: The disc diffusion method was used to determine the antimicrobial activity of *Aloe vera* juice (Barry and Thornsberry, 1985). Bacteria strains were cultured in trypticase soy broth (Difco) at 37°C for 24 h and reproduced as to contain 10^8-10^9 CFU mL^{-1} levels of organism. A volume of 0.1 mL of these cultures was inoculated on Mueller-Hinton agar (Oxoid) and spread on the entire surface of the dish using a sterile drygalski spatula. Then, sterile paper discs (Whatman: 1.6 mm) with absorbed *Aloe vera* juice (30 μL $disc^{-1}$) were placed onto the agar at certain intervals by pressing gently. After the plates were incubated at 35°C for 48 h, the inhibition zones around the discs where no growth occurred were measured in millimeters. *C. albicans* strain was inoculated in saboraud dextrose broth (Difco) and incubated at 25°C for 24 h and proliferated to 10^7 CFU mL^{-1} microorganisms. The 0.1 mL of prepared culture was transferred to saboraud dextrose agar (Difco) and above process was performed similarly. After this, these plates were incubated at 25°C for 24 h. At the end of the incubation period inhibition zone diameters, which were formed around the discs, were measured as millimeters. The screening was performed in duplicate.

RESULTS

The antimicrobial test results of *Aloe vera* juice are shown in Fig. 1. At the end of the analysis, *Aloe vera* had only antimicrobial activity on *M. smegmatis, K. pneumoniae, E. faecalis, M. luteus, C. albicans* and *B. sphericus*, but had no inhibitory effect on *P. aeruginosa, S. aureus, E. coli* and *S. typhimurium*. The least inhibitory effect was found on *M. luteus,* while *C. albicans* was detected to be the most sensitive strain.

In general, *Aloe vera* juice had inhibitory activity against Gram-positive bacteria with exception of *S. aureus*; but had no inhibitory activity at the proliferation of Gram negative bacteria with exception of *K. pneumoniae*.

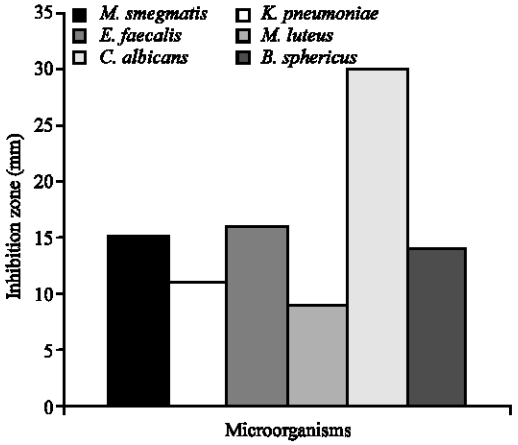

Fig. 1: Antimicrobial activities of *Aloe vera* juice

DISCUSSION

Numerous aloe species around the world are used for conditions ranging from dermatitis to cancer (Kemper and Chiou, 1999). Furthermore, the fresh gel, juice or formulated products have been used for medical, food and cosmetic purposes, as well as for general health (Park and Jo, 2006).

In the last decade, *Aloe vera* has been used extensively in health drinks, topical creams, toiletries and cosmetics (Anonymous, 2008) and there are many reported claims of its beneficial properties, encompassing a broad range of conditions (Shelton, 1991; Reynolds and Dweck, 1999).

There is a wide range of research from all over the world based upon different species of Aloe for antimicrobial activity. In previous studies, it was indicated that the isolates from *Aloe vera* were shown to inhibit microbes like *Staphylococcus* sp. *S. aureus* (Lorenzetti *et al*., 1964; Heggers *et al*., 1979; Martínez *et al*., 1996; Agarry *et al*., 2005; Cete *et al*., 2005; Kaithwas *et al*., 2008), *Candida* sp. *C. albicans* (Heggers *et al*., 1979; Stuart *et al*., 1997; Agarry *et al*., 2005; Cete *et al*., 2005), *P. aeruginosa* (Soeda *et al*., 1966; Agarry *et al*., 2005; Cete *et al*., 2005; Kaithwas *et al*., 2008), *Klebsiella* sp./*K. pneumoniae* (Heggers *et al*., 1979; Heck *et al*., 1981), *E. coli* (Lorenzetti *et al*., 1964; Heggers *et al*., 1979; Cete *et al*., 2005; Kaithwas *et al*., 2008), *M. luteus* (Cete *et al*., 2005) and *E. faecalis* (Robson *et al*., 1982; Kaithwas *et al*., 2008).

In the study, *Aloe vera* juice had an inhibitory effect at different levels against *M. smegmatis, K. pneumoniae, E. faecalis, M. luteus, C. albicans* and *B. sphericus*. However, a similar effect against other microorganisms (*P. aeruginosa, S. aureus, E. coli* and *S. typhimurium*) was not found. According to the results of the tests, it

can be suggested that *C. albicans* (inhibition zone: 30 mm) was the most sensitive strain to *Aloe vera* juice and *E. faecalis* (16 mm), *M. smegmatis* (15 mm) and *B. sphericus* (14 mm), respectively. The weakest inhibitory effect was found against *K. pneumoniae* (11 mm) and *M. luteus* (9 mm).

The antibacterial activity of the plant juice was found mainly against the Gram-positive bacteria. *Aloe vera* juice was the only one to show activity against *K. pneumoniae*, a Gram-negative bacterium. These differences may be attributed to the fact that the cell wall in Gram-positive bacteria consists of a single layer, whereas the Gram-negative cell wall is a multi-layered structure and quite complex (Matu and Van Staden, 2003).

Reports have mainly focused on the antidiabetic, anticancer and antimicrobial properties of the whole leaf, gel, or juice of the plant. Whole-leaf components proposed to have direct antibacterial properties include anthraquinones (Anonymous, 2008) and saponins (Reynolds and Dweck, 1999; Urch, 1999), while polysaccharides have been attributed with indirect bactericidal activity through stimulation of phagocytic leukocytes to destroy bacteria (Lawless and Allan, 2000; Pugh *et al*., 2001).

Anthraquinones contain phenolic compounds which are found exclusively in the plant sap. The important ones, aloin, aloe-emodin and barbaloin, act as painkillers. They also function as antibacterials and antivirals (Shelton, 1991). It was also, reported to have anti-inflammatory activity (Yamamoto *et al*., 1991) and genetic toxicity (Muller *et al*., 1996). Saponins are capable of cleansing and having antiseptic properties. They act powerfully as antimicrobials against bacteria, viruses, fungi and yeasts (Anonymous, 2008).

Scientific evidence has brought about the possibility of the utilization of plant extracts in the treatment of fungal and bacterial infections and the development of antibacterial and antifungal products (Farnsworth, 1984). Furthermore, antibacterial activity has also made a better understanding of the use of traditional medicines as potential drugs in addition to contemporary drugs (Coopoosamy and Magwa, 2007).

CONCLUSION

It was determined that *Aloe vera* juice had inhibitory effect against pathogenic bacteria, causing food poisoning or different diseases in humans, especially Gram-positive bacteria. We think that it can be alternative to chemicals used in medication, food and cosmetic sectors.

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