JOURNAL OF
EMERGING INVESTIGATORS

Article

# Are alkaline spices the future of antibiotics?

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## SUMMARY

**Some developing countries do not have access to the same medicine and antibiotics that developed countries do. Thus, we chose to focus on compounds and spices that have antimicrobial properties with a history of being used in traditional medicine but are not commonly used in developed countries. We experimented with the most commonly available alkaline spices (turmeric, cayenne pepper, and cinnamon) to study their antimicrobial properties. Our hypothesis was that alkaline spices would have antimicrobial activity. We used oral swabs from three volunteer subjects to inoculated blood-agar plates. The result of our 24-hour (bacteriostatic) experiment showed a zone of inhibition of bacterial growth, with the largest zone of inhibition being around turmeric, followed by cayenne pepper, and the smallest around cinnamon. The 48-hour (bactericidal) results were similar, but with added complexity in turmeric and cayenne pepper showing a mold-like substance with a zone of bacterial inhibition not seen with cinnamon. These results are impactful and support our hypothesis, as alkaline spices generally do show antibacterial properties and both bacteriostatic and bactericidal effects correlated with degree of alkalinity. Further studies with an increased number of spices and with a larger sample size are needed to validate our findings.**

## INTRODUCTION

The Sushruta Samhita (सुश्रुतसंहिता, IAST: Suśrutasaṃhitā, literally “Suśruta’s Compendium”) is an ancient Sanskrit text on medicine and surgery, and one of the most important such treatises to survive from the ancient world, which focuses on preventing diseases and curative remedial procedures (1-2). The Sushruta Samhita, along with the Sanskrit medicine-related classics Atharvaveda and Charak Samhita, describe more than 700 medicinal herbs (3). In these ancient books, curcumin – the active ingredient in the spice turmeric – was described as an anti-infective, anti-inflammatory, and antidiabetic. Scientists are following intriguing leads that curcumin might have antimalarial activity and inhibit the replication of some viruses (3).

Ayurveda (the ‘science of life’) is a system of traditional medicine native to India and practiced in other parts of the world as a form of alternative medicine (4). Ayurveda stresses using hundreds of vegetable-derived compounds, including those found in turmeric, cinnamon, and cayenne peppers (5). Cayenne pepper is known in Ayurvedic medicine to increase internal heat and has been traditionally used for respiratory diseases, as well as conditions related to the intestinal microbiota (6). Cayenne pepper features cleansing and antioxidant properties and enhances bioavailability, meaning it helps the body absorb and circulate needed nutrients (6). Additionally, cayenne pepper may help promote free flow of oxygen to the brain, support digestion and circulation, stimulate appetite, and support joint health (7). The antidiarrheal activity of cinnamon is also well-documented (8). The ‘Indian Materia Medica’ and the ‘Indian Medicinal Plants – A Compendium of 500 species’ classifies cinnamon as an herbal drug having cardiovascular effects (9-11).

In this investigation, we wish to explore the applicability of Ayurvedic compounds in reducing common ailments. As alkaline spices are taken orally, oral diseases represent a clinical setting in which to explore the benefit of these compounds. Oral diseases are primarily caused by bacterial infections (12). The oral cavity is comprised of many surfaces, each coated with a plethora of bacteria, the proverbial bacterial biofilm (13). Some of these bacteria have been implicated in oral diseases such as tooth decay and periodontitis, which are among the most common bacterial infections in humans (14). One key environmental factor affecting the pathogenicity of oral biofilms is pH, with evidence mounting that alkali generation plays a major role in moderating the progression of dental decay (15).

Given the ready availability of turmeric, cayenne pepper, and cinnamon and their role previously documented in ancient medicine, we decided to study the effects of these three compounds on the growth of nonpathogenic oral bacteria. Furthermore, as these three spices are alkaline in nature (turmeric is -46.66 on the alkalinity scale; cayenne pepper is -31.44; cinnamon is -23.75, with a lower number being more alkaline) (16), we hypothesized that these spices would inhibit bacterial growth.

## RESULTS

To test whether alkaline spices inhibit bacterial growth, we used oral swabs from three subjects to inoculate blood agar plates. A droplet of each spice mixed in water was placed in the center of each plate. We recorded the size of the zone of inhibition around each spice after 24 and 48 hours of growth at 35°C. After 24 hours, each subject's plate showed a decreased area of bacterial colonies around turmeric (approximately 5mm), cayenne pepper (approximately 3mm), and cinnamon (approximately 1mm) as measured by the zone of inhibition around the spice droplet (**Figure 1**). When diluted samples of each of the spices were used, there was still a zone of inhibition around each spice droplet, and qualitatively turmeric had the largest zone followed by cayenne pepper and then by cinnamon (data not shown).

We then prolonged the incubation to 48 hours and there was a mold-like substance growing on the blood agar plate which was not seen in the control but was seen in the plates containing turmeric and cayenne pepper, but not the cinnamon. In this mold-like area the bacterial growth was inhibited (**Figure 2**). As with the results at 24 hours, diluted samples of the spices were used and resulted in similar outcomes as the undiluted samples.

## DISCUSSION

Antibiotics are a class of compounds that can be used to treat patients with bacterial infections – most antibiotics target bacterial functions or growth processes (17). Although these drugs are largely effective, the widespread use of antibiotics has led to the development of antibiotic-resistant bacteria (17). Natural products (such as alkaline spices) are a major component of most Eastern diets, and have multiple host cell-protective effects, including anti-inflammatory, anti-viral, anti-fungal, anti-bacterial, antioxidant, anti-arthritis, and memory/cognitive function enhancement (18). There are dense populations of pathogenic and non-pathogenic bacteria in the oral cavity. Recently, studies have found a link between periodontal pathogens, gum hygiene, and Alzheimer's disease (18). Oral hygiene is associated with many other diseases including certain cancers (19). We hypothesized that since these non-pathogen bacteria are present in the oral cavity, natural substances that are widely available might inhibit their growth.

Generally, our 24-hour results support our hypothesis that alkaline spices have bacteriostatic properties, and the more alkaline a spice was, the more bacteriostatic it was (turmeric > cayenne pepper > cinnamon). Our 48-hour results, which were done to examine bactericidal properties of alkaline spices, were somewhat more complex. Specifically, bactericidal properties related in part to the formation of a mold-like substance (similar perhaps to penicillin) seen in the dishes with spices but not the control (20). Whereas turmeric and cayenne pepper both had this mold-like substance with bactericidal properties, cinnamon did not. However, there was general correlation between the alkalinity of the spice and the zone of inhibition (again, turmeric > cayenne pepper > cinnamon).

Our study does have limitations. First, the sample size, in terms of (a) the number of spices studied, (b) the number of plates incubated, and (c) the number of volunteers (all three of whom were high-school aged males), was small/limited, so a statistical analysis could not be done. Second, the origin of the mold-like substance seen in the 48-hour experiments is not clear and may perhaps be related some other property of the spice not related to the alkalinity. However, possible contamination is difficult to rule out, as the controls were not swabbed with sterile saline, but it should be noted that there was no growth of this mold-like substance in the control group.

Within these stated limitations, however, our study does provide some initial support for the hypothesis that alkaline spices may help with oral hygiene by anti-bacterial action. Particularly as oral hygiene has been linked with serious

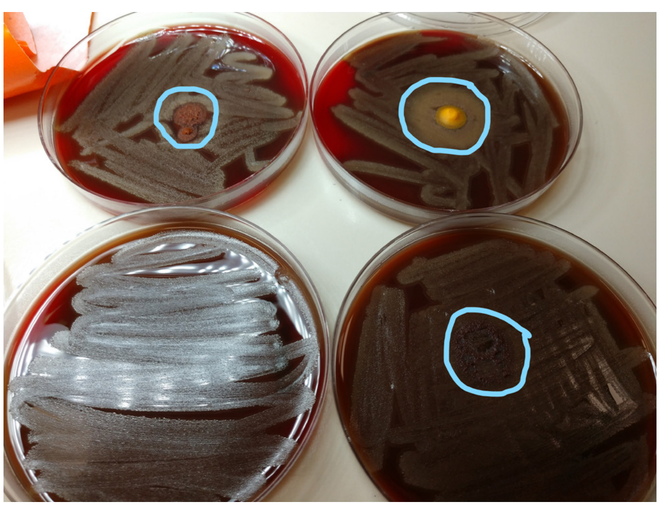

**Figure 1. 24 hour (bacteriostatic) incubation results with zone of inhibition circled.** Clockwise from lower left: control (no zone of inhibition), cayenne pepper (~3mm), turmeric (~5mm), and cinnamon (~1mm).

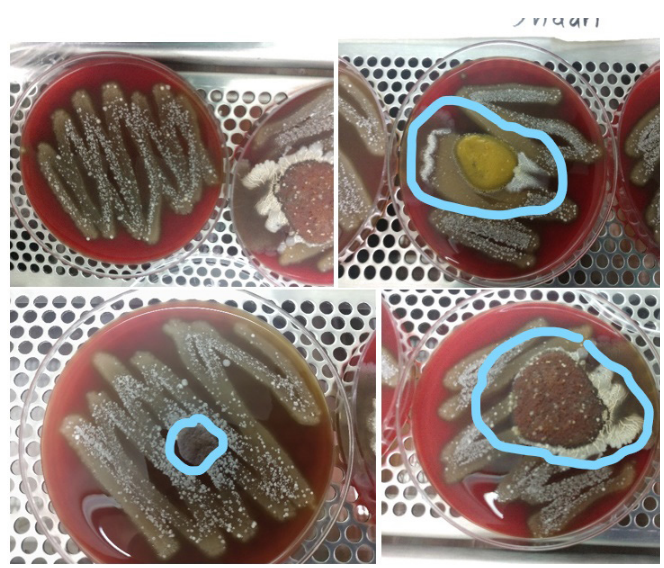

**Figure 2. 48 hour (bactericidal) incubation results.** Clockwise from upper left: control (no mold), turmeric (mold in zone of inhibition), cayenne pepper (mold in zone of inhibition), and cinnamon (no mold/ no zone of inhibition).

diseases such as cancer and Alzheimer's disease, alkaline spices may have a role to play in preventing these ailments (18,19). Alkaline spices may help decrease antibiotic resistance and improve oral hygiene using a natural approach that has potentially fewer side effects and lower cost compared with antibiotics. More work is needed to determine whether an individual's normal diet can influence oral hygiene.

Further studies, both with a larger range of spices and with a larger sample size, are needed to validate our bacteriostatic and bactericidal results. Future efforts should also include exploration of the nature of the mold-like substance we observed.

## MATERIALS AND METHODS

### Media and spice preparation

Blood-agar plates were prepared using standard procedures. Spices were acquired from a grocery store (Swad® brand). Sterilization of the spices, water and metal spoons was carried out for 1 minute using humid heat via an Instant Pot™ (**Figure 3A**) (21). Then we dissolved 1 gram of each spice in the sterilized water. To achieve the same level of semisolid consistency for each spice, we used 4.5 mL of water to dissolve the turmeric, 3.0 mL of water to dissolve the cayenne pepper, and 2.5 mL of water to dissolve the cinnamon (**Figure 3B**). The 'concentration' of turmeric was the lowest, followed by cayenne pepper, followed by cinnamon. Diluted concentrations of spices were made by adding more water by 1 mL for each spice (in total, 5.5 mL of water to dissolve of turmeric, 4.0 mL of water to dissolve cayenne pepper, and 3.5 mL of water to dissolve the cinnamon). Then, we added 2 more milliliters of water to dissolve the spice (6.5 mL of water to dissolve of turmeric, 5.0 mL of water to dissolve cayenne pepper, and 4.5 mL of water to dissolve the cinnamon).

### Inoculation and incubation

Each volunteer subject first gargled their mouth with tap water, and then used a sterilized swab to swab their own buccal cavity, oral mucosa, and tongue. Then, we streaked each plate with the swabs containing the oral bacteria. We dropped the dissolved spice in the center of the plate with a sterilized inoculating loop (0.1 mL of each spice) to achieve uniform coverage.

We incubated all test blood agar plates with the spices at 35°C for 24 hours, recorded the results (for the primary solution concentrations only), and incubated further for a total of 48 hours When we recorded the results to see the amount of bacterial growth on each individual spice at each time point (24 or 48 hours). For the primary solution concentration (first series described above) at 24 hours only, we specifically measured the zone of inhibition (from the edge of each spice) in mm with a ruler.

**Received:** November 11, 2021
**Accepted:** January 8, 2022
**Published:** January 23, 2022


**Figure 3. Experimental setup. A)** Sterilization setup in the Instant Pot™. **B)** Uniform semisolid consistency of each spice prior to plating.

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