

# [Free antimicrobial effect of lemon, garlic and whole jalapeno on bacillus in a di...](https://assignbuster.com/free-antimicrobial-effect-of-lemon-garlic-and-whole-jalapeno-on-bacillus-in-a-disk-diffusion-report-sample/)

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## Lab report #

Introduction   
In recent times, synthetic antibiotics have caused resistance in microorganisms and also, have shown unwanted side-effects. This has demanded for alternate therapies to combat infections. Scientists have developed interest in plant extracts, especially the ones that are traditionally used as spices, for new antimicrobial agents. Spices are traditionally used to give a distinctive flavor to the food but, they also contain potential antimicrobials which are also documented in literature. Even Louis Pasteur demonstrated the antibacterial effect of garlic and onion extracts. Salsa contains three main ingredients which have potential antimicrobial compounds. First, Lemon (Citrus Limon) is reported to contain antimicrobial Limonene and Citral that are the major component of essential oils (EOs), while Linalool, a terpene determines the strength of the fragrance (Fisher 1233). Garlic (Allium Sativum), another spice of salsa, contains sulfur containing thiosulfinates and allicin compounds, which are also reported to show antimicrobial activity (Tamara 3). Jalapeno (Capsicum annuum) pepper in salsa contains capsaicin which is also supposed to show antimicrobial activity (Tamara 3). These are also used in many food items as preservatives. Fisher and Phillips have shown the antimicrobial effect of lemon essential oils on the growth of B. Cereus with a zone of about 18 mm. Durairaj et al. studied the effect of aqueous extract of garlic on various bacterial species including B. Subtilis. They found a zone of inhibiton ranging from 15-54 mm at different concentration, with the minimum effective concentration only between 7-21 mgmL. Tamara and Marsh have demonstrated that at a concentration of 500 mg/mL, Jalapeno extract failed to show antimicrobial effect on B. Cereus and was slightly effective on other species including gram-positive and negative organism. Thus, if the above mentioned spices do have antimicrobial compounds, it would be interesting to see its effect on the growth of bacteria. Thus, we decided to study the extracts of these three spices on the growth of Bacillus, common food contaminant bacteria. We assume that lemon juice will show maximum inhibition as it has more acid content. Many workers have worked on the effect of different spices on bacterial growth.

## Method

The spices in question were first extracted for the potential antimicrobial compounds. The Lemon was peeled and cleaned with sterile water, later the juice was squeezed in a sterile glass tube. The juice was weighted and diluted with sterile water to give the final concentration of 4 mg/mL. Garlic and jalapeno were also cleaned with sterile water and placed in clean and dry motor. They were crushed and sterile water was added to the motor so that the concentration of the extract reached 4 mg/mL. The technique used to determine the antimicrobial activity of the extract was disk diffusion assay. A day before the assay, the Bacillus was transferred from the preserved agar vial in to a nutrient broth and kept for overnight incubation so that active bacteria are available for the assay. In brief, in a sterile nutrient agar plate, 0. 1 mL of the turbid suspension was aseptically placed in the center of the plate. The suspension was spread over the entire surface of the plate to give a lawn like growth. The plates were set aside and then the appropriate numbers of sterile disks each measuring 7 mm were dipped in the previously prepared extracts. In one plate, two halves were made; in one half a control disk dipped in sterile water was placed and in the second half the disk dipped in the respective extract was placed. The plates were incubated for 24 hrs at 37 degree Celsius. After the incubation, the plates were looked for a lawn like growth on the surface. The area around the disk dipped in extract was checked for an inhibition and the diameter of the zone was measured, in mm, using a standard ruler.

## Results

Observation showed that after incubation, the growth of Bacillus was not lawn-like and lacked visible proof of growth. The control disk and the disks soaked in the extract did not show any inhibition and in-fact showed some growth around them. The surface of the plates also showed isolated colonies of microorganisms.   
Figure: Effect of Lemon (A), Jalapeno (B) and Lemon (C) on the growth of Bacillus

## Conclusion

The antimicrobial effect of the three extracts was inconclusive and needs repeat experiments to prove it. There were many errors encountered during the process of conducting the assay. The suspension did not contain enough growth of Bacillus to form a lawn over the agar surface. The method and technique used was not aseptic and as a result there were isolated colonies of contaminants seen on the surface of the plate. Also, the water or diluents used for soaking the disks was contaminated; there was growth of microorganisms around the disks. Lastly, the concentration of 4 mg/mL may not be sufficient to show any noticeable effect on Bacillus. The hypothesis that lemon, garlic and Jalapeno have antimicrobial compounds could not be established due to erroneous techniques and low concentration of extracts. It is recommended to repeat the assay using proper sterile glasswares, following aseptic techniques and higher concentrations of the extracts.

## References

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