Antimicrobial properties of
*Morinda citrifolia* (Noni) root

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Abstract
The purpose of this project is to determine whether the Noni (Morinda citrifolia) root contains antimicrobial properties. A traditional extraction method was used to obtain the juice from the Noni root, which involves the scrapping of the cortex, pounding and straining of Noni root juice using cheesecloth. The Kirby Bauer standard antibacterial testing assay was used to test the root extract against four bacterial strains (Bacillus subtilis, Escherichia coli, Staphylococcus aureus, and Pseudomonas aeruginosa) and the yeast, Candida albicans. The resulting zones of inhibition observed formation of distinct zones of inhibition with B. subtilis, S. aureus, and E. coli but no inhibition with P. aeruginosa and C. albicans. The ANOVA Tukey test confirmed that the zones of inhibition provided by pure root extract was significantly more effective than 80% extract and the negative controls but less effective than the positive controls ($q > 4.23$ at $p = 0.05$). The test also confirmed that 40 µL dosages were more effective than the 20 µL & 30 µL.

Introduction
Herbal medicine has provided a mode of healing in a wide range of ailments and has been shown to be a cheap and effective source of medicine (Alan 1999). Locher et al. (1995) noted that Polynesians had used a variety of plants for treating infectious diseases. However, most Pacific island medicinal plants such as Centella asiatica (L), Calophyllum inophyllum (L) and Solanum nigrum L have yet to be studied due to geographical isolation from the western world. Their potential therapeutic properties are still unknown (McClatchy 2002, Steiner 1986).

Morinda citrifolia, a plant commonly used for its therapeutic properties through out the South Pacific, is a small shrub, three to twelve meters high (Wang, Su 2001). M. citrifolia is commonly known in various cultures as Indian Mulberry, Ba Ji Tian, Nono, Nonu or Noni, Cheese Fruit and Nhau (Wang et al. 2002). Noni belongs to the family Rubiaceae in the genus Morinda which has about 80 species of old world origin (Morton 1992). It is originated from Southeast Asia and spread via oceans currents, animals, birds and through human activities. It commonly found along the open coastal regions from sea level to the forested areas of 1300 meters. Polynesians have been utilizing the different parts of Noni for curing several tropical ailments (Locher et al. 1995). Its fruit and leaves have been used for treating arthritis, diabetes, high blood pressure, menstrual difficulties, boils, tuberculosis, broken bones, deep cuts and sores.
(Wang et al. 2002, Dasilva et al. 2004). Even now, however, its activities remain obscure to science (Nostro et al. 2000, McClatchy 2002). The Noni (M. citrifolia) therapeutic properties against several bacterial infections such as boils, tuberculosis and ulcer suggest that noni extract might contain antimicrobial agents. Previous studies found no antimicrobial properties in Noni leaves and fruit extracts (Tavana 2003 & Sokia no date). The purpose of this project is to determine whether the roots of the Noni contain antimicrobial properties against *B. subtilis*, *S. aureus*, *E. coli* and *P. aeruginosa* and *C. albican*.

**Methods**

Noni roots were obtained from Laie, Oahu, Hawaii. The roots were cut from the plant and cleaned under running tap water. The epidermis was removed and the cortex was scraped and collected as a sample.

It was then pounded, extracted by straining using cheese cloth and stored at 4 °C. The extract was subsequently divided into two equal portions. One was assayed in its pure form and the other was diluted to 80% with distilled water.

Each extract was assayed for antimicrobial activity by the Kirby-Bauer method (Bauer et al. 1966). *Bacillus subtilis*, *Escherichia coli*, *Staphylococcus aureus* and *Pseudomonas aeruginosa* were cultured on Tryptic Soy Agar, and *Candida albicans* was cultured on Yeast Peptone Dexticase (YPD) medium. The cultures were streaked onto the appropriate agar media using a cotton swab and incubated for 24 hours. The plates were dried for 3 to 5 minutes followed by the application of Noni root extract impregnated disks. Blank disks were impregnated with the appropriate doze of Noni root extract (20µL, 30µL and 40 µL). Positive control, erythromycin for *Bacillus subtilis*, tetracycline (30 µg) for *Staphlococcus aureus*, vancomycin (30 µg) for *Pseudomonas aeruginosa*, chloramphenicol (20 µg) for *Escherichia*
coli and 1% Tolnaftate Topical antifungal for Candida albican were used. The plates were incubated immediately for 24 hours at 35°C for bacterial strains and 30°C for fungal strain. Each dosage; was replicated 5 times for each strain. After incubation, the diameter of zone of inhibition was measured in millimeters across the disk. The ANOVA Tukey test was used to analyze the significance of the zones of inhibition obtained by both 80% and 100% Noni root extract in comparison to their positive controls.

**Results and Discussion**

All concentrations (80% & pure root extract) had antimicrobial activity against Staphlococcus aureus, Escherichia coli & Bacillus subtilis. The zones of inhibition obtained by pure extract was significantly more effective than the 80% extract and negative controls (q > 4.23 at p = 0.05) but less effective than positive controls. The positive controls (tetracycline, chloramphenicol & erythromycin) were the most effective compared to both pure and 80% diluted Noni root extracts. Negative controls (distilled water & blank disk) did not inhibit the growth of the 3 bacterial strains (Figure 1, 2 & 3). The ANOVA Tukey test (q = 4.23 at p = 0.05) confirmed that pure extract concentration was significantly more effective compared to the 80% concentration against S. aureus, E. coli and B. subtilis.
Dilution effect of 100%, 80%, positive control & negative controls against S. aureus

Figure 1. Concentration effect expressed as average diameter zones of inhibition with standard deviation against S. aureus.

Dilution effect of 100%, 80%, positive control & negative controls against E. coli

Figure 2. Concentration effect expressed as average diameter zones of inhibition with standard deviation against E. coli.
All dosages (20 µL, 30 µL & 40 µL) of both pure and 80% root extract had antimicrobial properties against *S. aureus*, *E. coli* & *B. subtilis*. The positive controls (Tetracycline, Chloramphenicol & Erythromycin) were the most effective compared to both pure and 80% diluted Noni root extracts. Negative controls (distilled water & blank disk) did not inhibit the growth of the 3 bacterial strains. All dosages (20 µL, 30 µL & 40 µL) of both pure and 80% root extract showed antimicrobial properties against *S. aureus*, *E. coli* & *B. subtilis*. At pure & 80% concentration, 40 µL dosage was significantly more effective than 20 µL & 30 µL dosages against the three bacterial strains (*q* > 4.23 at *p* = 0.05). There was little or no significant effect between 20 µL & 30 µL (Figure 4 & 5). The ANOVA Tukey test also confirmed that the higher the dosage, the more effective the Noni root extract is (40 µL > 30 µL > 20 µL).
Figure 4. Dosage effect expressed as average diameter zones of inhibition with standard deviation against different bacteria.

Figure 5. Dosage effect at 80% concentration expressed as average diameter zone of inhibition with standard deviation against different bacteria.
In conclusion, *Morinda citrifolia* root extract has antibacterial activity against *Staphlococcus aureus*, *Escherichia coli* and *Bacillus subtilis* but not against *Candida albicans* and *Pseudomonas. Aeruginosa*. Previous Senior Biology research found no antimicrobial properties in the extracts of both the Noni fruit and the leaves (Tavana 2003 & Sokia) but found in the root. The antimicrobial activity produced by Noni root suggested that unlike fruit and leaves, roots evolved a defense mechanism against insects, bacterial infections or different soil conditions. Possibilities for future studies may include testing Noni root extract against other fungal strains other than *C. albicans* and analyzing its organic components responsible for its antimicrobial properties.

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References cited


