# Chai Tea Promotes Ampicillin Susceptibility in MRSA

Hinsdale, Shannon<sup>1</sup>, Pace, Joel R<sup>1</sup>., Anderson, Emeli<sup>1</sup>, Favoreto, Silvio, PhD<sup>1</sup>

Cuesta College Biological Sciences Division, San Luis Obispo, CA, USA

### Abstract

Rationale: MRSA is a resistant, more aggressive strain of *S. aureus* that is presenting a challenge to both the public and medical communities. This can be seen in the 80,461 infections, coupled with 11,285 deaths, per year in the United States alone [4]. This crisis is costing the U. S. healthcare system up to \$9.7billion, or approximately \$60,000 per patient, annually [5]. Consequently, MRSA treatment requires more potent and expensive drugs, accompanied by longer hospitalization [6]. As can be deduced from the given information, our options for treatment are dwindling as cost are rising, both in mortality rates and financially. Resistance is an ongoing process that will require alternative intervention. In this work we tested a well known plant based product for the ability to modify the resistance profile in newly isolated MRSA strains.

Methods: Aqueous based chai infusion (CW) or ethanol based chai extract (CA) were added to the media in standard diffusion antimicrobial testing. MRSA was isolated from the nostrils of healthy carriers and characterized by culture and PCR.

**Results:** The aqueous based chai infusion and ethanol extract promoted a significant increase in MRSA susceptibility to Ampicillin, as shown by the diameter of the inhibition halo (p<0.05)

Conclusions: Our results indicate that the aqueous based chai infusion or extract potentiated the action of Ampicillin against several isolates of MRSA in vitro. Interestingly, only Ampicillin, from all tested antibiotics, had shown synergistic action with chai. Drawing from these results, we propose the integration of natural plant compounds, with what are currently ineffective antibiotics against resistant strains, as a means of modifying resistance in MRSA [4].

## Aims

- To determine the ability of chai tea to modify the antimicrobial resistance of MRSA "in vitro"
- •To address differences in efficiency between water-base and alcohol base extracts.

# Methods

#### Chai extracts

• Water-based and alcohol-based extracts were prepared by infusion of cinnamon, cloves, cardamom, ginger, star anise, black tea and black peppercorn in hot water for one hour or in ethanol for a week. The extracts were filtered, bottled and kept at 4C. All experiments were performed with fresh batches of extracts, which were prepared weekly. The extracts were added to the media for standard disk diffusion antibiograms.

#### **MRSA** isolates

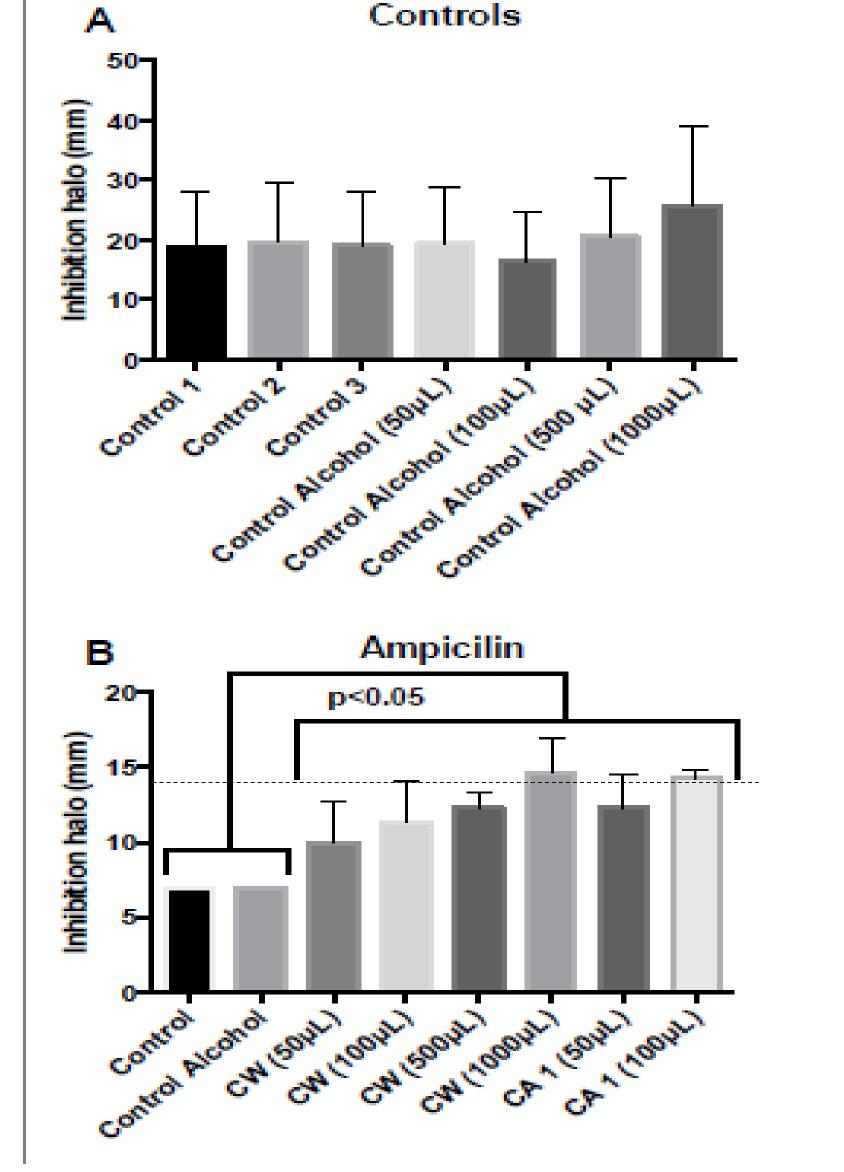
• We utilized 7 isolates of MRSA obtained from nasal swab survey of 260 college students. The swabs were used in Chromagar MRSA plates (Hardy Diagnostics) for initial isolation and the isolates identification confirmed by multiplex PCR assay using specific sequences for Staphylococcus ribosomes, Mec A and Paton-Valentine toxin.

#### Antibiogram

•We performed disk diffusion tests following NCCLS/CLSI standards. Extracts were added to the media before plate pouring. Prepared plates were used for tests within 12 hours. All tests were performed in quintuplicates.

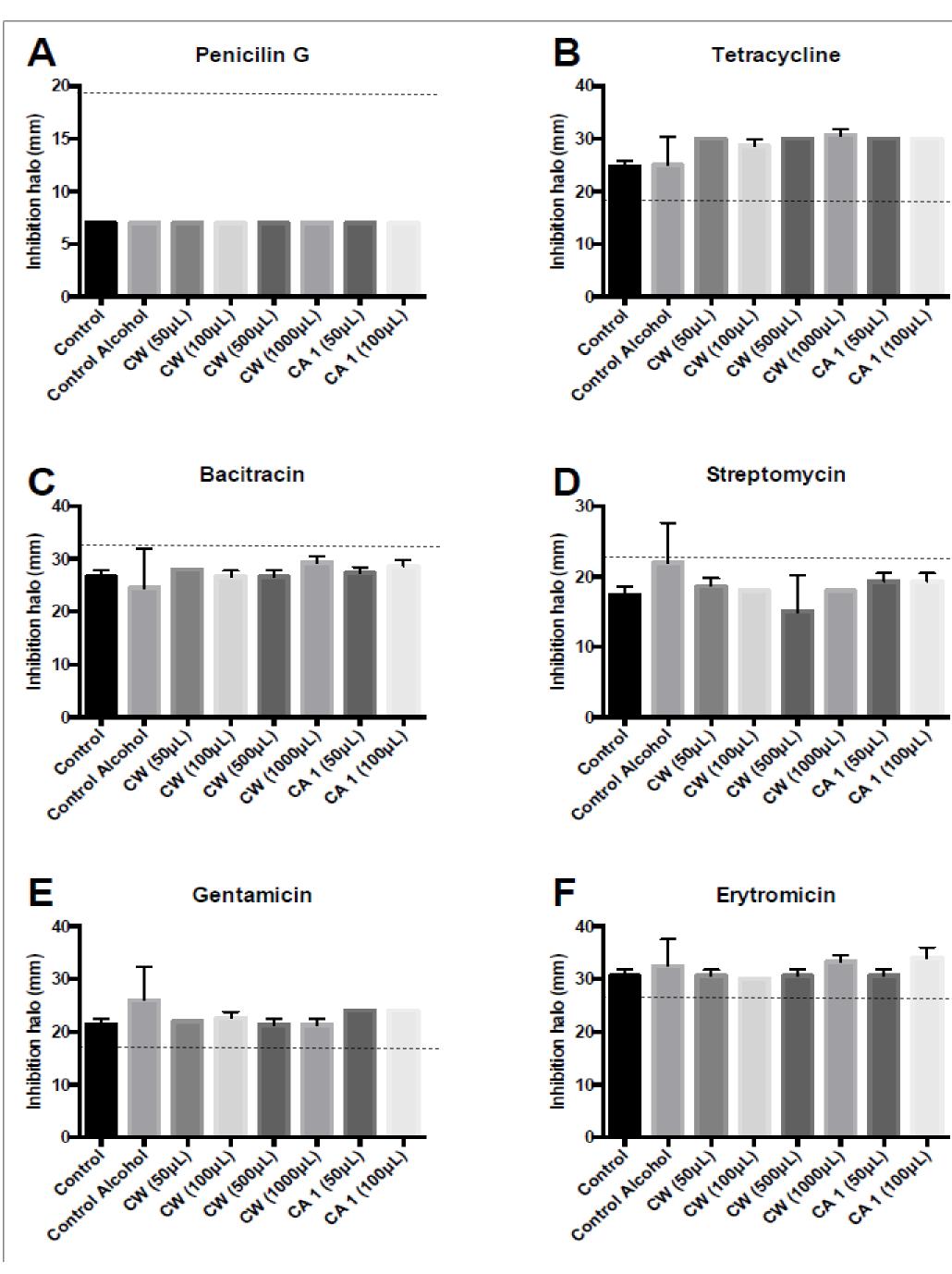
# Results

**Figure 1**—Inhibitory halo measurement in radial antimicrobial diffusion test.



A – Vehicle control test. B - Chai infusion (CW) or Chai ethanol extract (CA) were added to the media in controlled volumes. Results show average response of resistance to Ampicillin in seven MRSA isolates. Alcohol-based chai extract inhibited microbial growth at 500 and 1000µL and were not tested in antimicrobial resistance assays.

Figure 2—Inhibitory halo measurement in radial antimicrobial diffusion test



No significant synergistic effect between tested antimicrobial drugs and Chai infusion (CW) or extract (CA) added in controlled volumes. Results indicate that the synergistic effect on resistance is restricted to Ampicillin, among the tested antimicrobials.

# Summary

Our results indicate that the aqueous based chai infusion or extract potentiated the action of Ampicillin against several isolates of MRSA in vitro. Interestingly, only Ampicillin, from all tested antibiotics, had shown synergistic action with chai. Drawing from these results, we propose the integration of natural plant compounds, with what are currently ineffective antibiotics against resistant strains, as a means of modifying resistance in MRSA. Bacterial acquisition of resistance factors is not going to subside, it's an innate aspect of bacterial life being enhanced by extensive use of antimicrobial drugs[5,6,7]. Subsequently, the synergism shown between chai tea and ampicillin holds the possibility of assisting us in deconstructing mechanisms of resistance, helping stage for future studies to progress towards reducing our global crisis.

## Conclusion

- 1. <a href="http://www.emedicinehealth.com/mrsa">http://www.emedicinehealth.com/mrsa</a> infection/article em.htm#mrsa infection overview
- 2. Biggest Threats. (2016). Retrieved November 23, 2016, from <a href="http://www.cdc.gov/drugresistance/biggest\_threats.html">http://www.cdc.gov/drugresistance/biggest\_threats.html</a>
- MRSA The Superbug Poised to Cost Hospitals Super Sums. (2015, December 29). Retrieved November 23, 2016, from <a href="http://www.infectioncontroltoday.com/news/2015/12/mrsa-the-superbug-poised-to-">http://www.infectioncontroltoday.com/news/2015/12/mrsa-the-superbug-poised-to-</a>
- nttp://www.infectioncontroltoday.com/news/2015/12/mrsa-tne-superbug-poised-tocost-hospitals-super-sums.aspx
- 4. Cantley, A. (2016, January 25). Antibiotic Resistance: Old genes, new problems Science in the News. Retrieved December 06, 2016, from http://sitn.hms.harvard.edu/flash/2016/antibiotic-resistance-old-genes-new-problems/
- 5. Santiago, C., Pang, E., Lim, K., Loh, H., & Ting, K., (2014, June 30). Reversal of Ampicillin Resistance in MRSA via Inhibition of Penicillin-Binding Protein 2a by *Acalypha wilkesiana*. BioMed Research International, volume 14, Article ID 965348
- 6. Stevens, C., Rosado, H., Harvey, R., & Taylor, P.\*, (2015, September 28). Epicatechin gallate, a naturally occurring polyphenal, alters the course of beta-lactam-resistant Staphylococcus *aureus* in the zebrafish embryo. Frontiers in Microbiology, doi:10.3389/fmicb.2015.01043
- 7. Lahmar A, Bedoui A, Mokdad-Bzeouich I, Dhaouifi Z, Kalboussi Z, Cheraif I, Ghedira K, Chekir-Ghedira L, Reversal of resistance in bacteria underlies synergistic effect of essential oils with conventional antibiotics, *Microbial Pathogenessis* (2016), doi:10.1016/j.micpath.2016.10.18