

A model to study the inhibition of bacterial contamination of vegetable sprouts by plant based polyphenols

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Introduction

Consumption of vegetable sprouts is popular worldwide due to their nutritional value. However, several outbreaks of food borne diseases associated with sprouts have resulted in a decrease in sprout consumption in some countries. One of the main causes of food-borne diseases is contamination by enteric pathogens such as *Escherichia coli*. We used plant based bioactives to decrease bacterial adhesion and therefore contamination using a sprouted legume (alfalfa) model.

Materials and Methods

Bacterial adhesion in sprouted food model¹

Three day old alfalfa sprouts were inoculated with high and low counts of *Escherichia coli* (Environmental science and Research # 916) for 4 h in a 25 $^{\circ}$ C incubator on a rotating shaker at 40 rpm. For microbial analysis, 10 sprouts were homogenized with phosphate buffered saline and adherent bacteria were counted by standard dilution and plating techniques.

Antibacterial screening of polyphenols²

We screened a range of polyphenols to identify the compound with maximum antibacterial activity. Broth microdilution assay was carried out to identify the maximum inhibitory concentration (MIC) of polyphenols, i.e. the lowest concentration of an antibacterial compound required to inhibit the growth of bacteria $(1.5x10^7$ cfu/ml) after incubation for 18 h.

Polyphenol treatment of E. coli inoculated alfalfa sprouts

Bacterially contaminated sprouts were washed with gallic acid (3 mg/ml) for 5 min and microbial adhesion was assayed by standard dilution and plating techniques.

Results

- Gallic acid was the most effective plant based polyphenol against *E. coli* (Table 1).
- Treatment with gallic acid in the contaminated sprouts (Figure 1) inhibited attachment of bacteria by about 60 %.
- Gallic acid treatment appeared to decrease bacterial biofilm polysaccharide matrix in the contaminated sprouts (Figure 2).

Table1. Minimum inhibitory concentration (MIC) of polyphenols for *E. coli*.

Polyphenols	MIC (mg/ml)
Quercetin	>5
Ellagic acid	>5
Chlorogenic acid	>5
Ellagitannin	>5
Gallic acid	3
Ferulic acid	>5
Epicatechin	>5
Rutin	>5
Catechin	>5



1.Barak, J. D., L. C. Whitehand and A. O. Charkowski (2002). "Differences in attachment of Salmonella enterica serovars and Escherichia coli 0157 : H7 to alfalfa sprouts." <u>Applied and Environmental Microbiology</u> 68(10): 4758-4763.

2.Camporese, A., et al. (2003). "Screening of anti-bacterial activity of medicinal plants from Belize (Central America)." Journal of Ethnopharmacology 87(1): 103-107









Figure 2. Scanning electron images of alfalfa sprouts inoculated with *E. coli* followed by (a) no treatment and (b) gallic acid treatment.

Conclusion

The results of our study suggest that the plant based polyphenols may provide an effective, natural intervention strategy to retard bacterial adhesion.

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