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Nasturtium antimicrobial activity against *Streptococcus agalactiae*

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Abstract
This work uncovered the antibacterial properties of *Nasturtium* against gram-positive *Streptococcus agalactiae*. Certain strains of *Streptococcus* have demonstrated antibiotic resistance. *Streptococcus agalactiae* commonly infects the elderly, neonates, and pregnant women, which makes this a pathogen of concern. Samples of Nasturtium leaves and flowers were extracted using 95% ethanol as extraction solvent and filtered. The filtrate was impregnated into sterile discs. Discs infused with extracts or extraction solvent as control, were placed on *S. agalactiae* plated plates. Clear zones around disc indicating bacterial susceptibility were measured. Nasturtium demonstrated antibacterial activity against *S. agalactiae* with a mean zone of inhibition of 20 mm. This suggest nasturtium can potentially be developed as an antibacterial agent against *S. agalactiae*.

Keywords: *Nasturtium*, *Streptococcus agalactiae*, neonates, and pregnant women, plated plates

Introduction
As microbes continue to evolve due to mutations, their resistance to antimicrobials is rapidly becoming one of the largest challenges of the future for human survival [1, 2]. Pathogens are quickly evolving to become multi-drug resistant in order to counteract the effects of antimicrobials used in clinical practice today [1]. Bacteria have used several mechanisms to develop antibiotic resistance, including cell wall modifications, alteration of actions of efflux pumps, and changes in gene expression [1, 2]. Antibiotic resistance has led not only to annual treatment cost increases in the billions of dollars, but a large increase in mortality for many patients [3,4]. Development of new antimicrobials must be undertaken to combat the growing resistance before certain pathogens become untreatable [5, 6].
Plants have been used by many people throughout the world to treat illness and disease [7, 8]. Numerous plants have demonstrated antimicrobial activity, and could potentially serve as the future of antimicrobials [7, 9]. *Tropaeolum*, better known as Nasturtium, is a plant that could potentially fulfill this role of a future antimicrobial. Nasturtium is native to South America and Mexico and is commonly found in homes, due to its pleasant smell and appearance [10]. Nasturtium is a plant with circular leaves and bright colored flowers [11]. Nasturtium plants grow rapidly and can withstand drought conditions [11]. In addition, Nasturtium is a plant that is edible for humans [11]. Previously, Nasturtium has shown strong antimicrobial activity against *Candida spp*. *H. influenzae, S. marcescens, M. catarrhalis, P. vulgaris, and* with high levels of susceptibility [12]. Nasturtium has also shown antimicrobial activity against *K. pneumoniae, S. aureus, S. pneumoniae, S. pyogenes, P. aeruginosa and E. coli* with intermediate susceptibility, and activity against *viridans enterococci* and *streptococci* with low susceptibilities [12]. While Nasturtium has demonstrated activity against many bacteria, the need remains to search for novel antimicrobial activity against other pathogenic bacteria. This experiment searches for this novel activity in nasturtium by testing it against *Streptococcus agalactiae.*
The Group B streptococcus, *Streptococcus agalactiae*, is a gram-positive coccus that is especially deleterious to expectant mothers, the elderly, and neonates. It is known to cause sepsis and preterm birth in neonates [13]. Consequently, *S. agalactiae* has been characterized as a pathogen of public health concern because it is implicated in the significant cause of morbidity and mortality in newborns [14, 15]. Prevention measures are vital to control this pathogen in this population [14]. *S. agalactiae* also often affects the immunocompromised, but has now begun infecting healthy people [15]. *S. agalactiae* previously demonstrated resistance to both beta lactam antibiotics (e.g. penicillin) and non-beta lactam antibiotics [13]. Identifying antibacterial agents against resistant and infectious strains of *S. agalactiae* is vital for future prevention and treatment [15].

Based on previously characterized antibacterial property of Nasturtium against various bacterial strains, we hypothesize in this experiment that Nasturtium demonstrate novel antibacterial activity against *S. agalactiae*.

Methods

2 grams of *Nasturtium* were extracted in 95% ethanol (Puga *et al*., 2022). Multiple paper discs were soaked in the filtrate or ethanol only (as solvent control) and air-dried (Puga *et al*., 2022). 100 ul of freshly cultured bacteria was diluted with 9 ml of 1% saline solution and 100 ul of this dilution was plated (Puga *et al*., 2022). The previously air-dried discs were placed on *S. agalactiae* plated plates and incubated at 37 °C s for 24 hours. Clear zones around discs were measured as a marker of bacterial susceptibility to the extract.

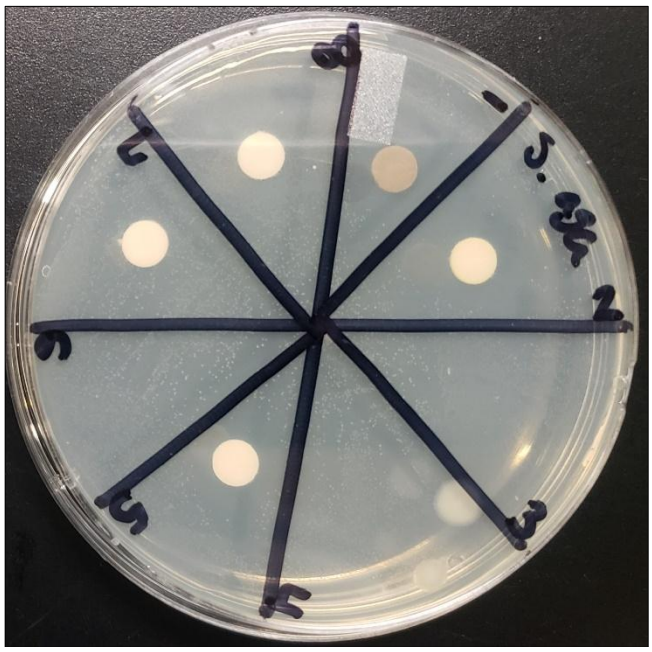

Fig 1: Nasturtium (Position 5) and blank disk (Positions 7, 8) against *S. agalactiae.*

Results

Nasturtium shows antibacterial activity against *S. agalactiae* with a mean zone of inhibition of 20mm.

Table 1: *Nasturtium*-infused disc induced zone of inhibition of *S. agalactiae.* Ethanol-infused disc (blank disc) induced no zone of inhibition of *S. agalactiae.*

	***S. agalactiae* (zone of inhibition, mm)**
Blank Disk	0
Nasturtium	20

Discussion

In this experiment, Nasturtium demonstrated novel activity against *S. agalactiae* with a mean zone of inhibition of 20 mm. *S. agalactiae* is a major pathogen of concern to the newborn population where it has shown large numbers of morbidity and mortality [14, 15]. *S. agalactiae* is also commonly infectious to pregnant women and the immunocompromised population [15]. Moreover, *S. agalactiae* is now demonstrating infections in people who are currently healthy [15]. The rise of infections in healthy people and the mortality in neonates is of significant concern when coupled with the fact that *S. agalactiae* has begun to demonstrate antibiotic resistance [13]. The novel activity of Nasturtium against *S. agalactiae* serves a good starting point for the development of an antibacterial for this pathogen. In addition, Nasturtium has previously demonstrated extensive coverage against other gram-positive and gram-negative bacteria [12]. This means that Nasturtium can be potentially developed as a broad-spectrum antimicrobial with coverage against both gram-negative and gram-positive bacteria. With the growing antibiotic resistance crisis, the search for compounds with antimicrobial activity is vital for the future of human health [1-6]. Based upon the novel results of this experiment and previous experiments, we believe there should be further investigation and subsequent development of Nasturtium as an antimicrobial agent.

Limitations of the Study: Resistant strains of *S. agalactiae* could not be specifically tested due to biohazard security concerns. We suggest further testing to be done in a biohazard secure facility, to determine susceptibility of resistant strains of *S. agalactiae* to nasturtium extract both *in vitro* and in *vivo*. Further testing should also be done to examine the efficacy and side effects of Nasturtium *in vivo*.

Conflicts of Interest

The authors declare no conflicts regarding this work.

Contributions of Authors

Conceptualization- PNA; Data curation-TP; Investigation-PLT, NI, PNA; Methodology-PNA; Manuscript writing- JS and TP; Visualization-JS; Review & editing- JS and PNA.

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Ethics approval statement

No ethics approval required.

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Resources

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