The Inhibitory Effects Demonstrated by Manuka Honey on Biofilms: How Manuka Honey May Soon Replace Conventional Antibiotic Therapy

Laura Pavelik, Assisted by Dr. Cundell, PhD
Philadelphia University, College of Science, Health, and Liberal Arts, Philadelphia, PA

Introduction

The surge of antibiotic-resistant bacteria has raised substantial concerns over how to effectively and efficiently control antibiotic-resistant bacteria capable of secreting biofilms. A biofilm can be defined as a self-producing extracellular matrix (Carter, et, al., 2016). Manuka honey, an alternative to conventional antibiotics, has proven successful in inhibiting planktonic cells and killing bacteria living under the protection of biofilms (Brudzynski & Sjaarda, 2015).

Formation of Biofilms

When the structures and functions of various biofilms were assessed, their mechanisms of resistance appeared to be dependent upon multicellular strategies. First, the higher the number of cells present in a given area increases the resistance of these cells to conventional antibiotics (Lin, et. al., 2010) In addition, biofilms contain persister cells, requiring minimal amounts of energy in order to survive for long durations (Cundell & Wilkinson, n.d.). Therefore, as the surface level cells die, they offer a protective layer of impenetrability for persister cells in the center of the biofilm (Cundell & Wilkinson, n.d.). Finally, in order for biofilms to be present, an initial adherence to a physical material is required. (Lin, et. al., 2010).

The Origins of Manuka Honey

Manuka honey is produced by selective Leptospermum species native to New Zealand and Australia, and is colonized by the honey bees, Apis mellifera (Brudzynski & Sjaarda, 2015). Levels of antimicrobial components contained in different batches of Manuka honey can range from 100ppm to greater than 1200ppm (Carter, et, al., 2016). Therefore, antibacterial activity of Manuka honey has to be tested individually. Medical-grade Manuka honey is subjected to gamma radiation in order to ensure its sterility (Carter, et. al., 2016). The medical community developed the phenol-equivalent scale as a standard to express the potency of antibacterial activity.

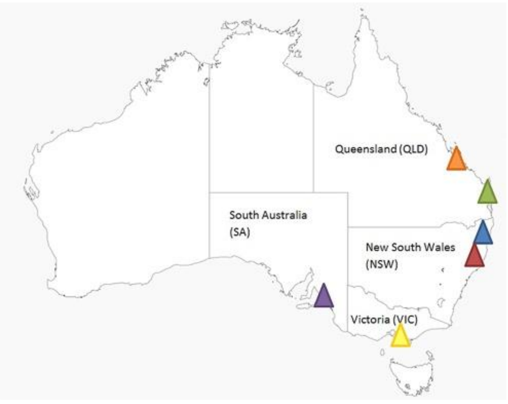


Figure 1: Areas where Manuka Honey is Prevalent

Figure 2: Medical-Grade Manuka Honey

Biofilms Susceptible to Manuka Honey

Methicillin-Resistant Staphyloccocus aureus (MRSA)

Jenkins and colleagues (2011) examined how Manuka honey successfully inhibits biofilm formation in MRSA and determined that Manuka honey was able to interrupt the cell cycle during the cytokinesis stage. Further, the research team noted a spike in the numbers of fully formed septa in MRSA, indicating that the bacteria was unable to successfully separate following the formation of cross walls (Jenkins, et. al., 2011). Henriques et. al. (2009) also confirmed this finding by detecting an increase in sensitivity of autolysin mutant, UspA, which is associated with bacterial cell division, and an essential component required for MRSA to exhibit full pathogenicity.

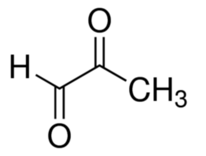
Clostridium difficile (C. diff)

With C. difficile contributing to 30-50% of all nosocomial infections, it is pertinent to determine and implement effective treatment methods in order to combat this species of bacteria (Giles, & Labeij, 2017). Currently, fecal microbiota transplantation (FMT) is the most widely accepted and implemented treatment method. Giles and Labeij (2017) cite the use of commercially available Manuka honey with UMF 26+ with a patient with persistent C. difficile infection. After treatment with two honey lavages a week apart, a sample removed from the patient's colon did not detect C. difficile (Giles, & Labeij, 2017). This discovery provides a promising alternative for the treatment of C. difficile infections.

Anti-Microbial Properties of Manuka Honey

Methylglyoxal: Unique Manuka Factor (UMF)

In foods and others honeys, methylglyoxal levels are usually between 3-37mg/kg and no greater than 24mg/kg respectively, whereas levels of methylglyoxal in Manuka honey have been recorded as high as 1541 mg/kg (Kwakman, & Zaat, 2012). Methylglyoxal successfully kills bacteria at submicromolar concentrations by a slow mechanism of action, explaining the potent but slow bacterial activity of Manuka honey. However, when just methylglyoxal was introduced to P. aeruginosa and E. coli samples, it did not prove to be completely inhibitory, suggesting other components may be partially responsible for antibacterial activity (Kwakman & Zaat, 2012).



Osmotic Stress, Leptosin, and Phenolic Components

Carter, et. al. (2016) successfully demonstrated that the combination of a high sugar content and low pH, which promotes osmotic stress, inhibits microbial growth, even when the honey has been diluted to 30-40% (Kwakman & Zaat, 2012). Methyl syringate 4-O-β-D gentiobiose, commonly referred to as Leptosin, a glycoside found exclusively in Leptospermum, was found to have a positive correlation with the UMF value. This may indicate that levels of leptosin may be linked to the modulation of antimicrobial activity (Carter, et, al., 2016). Marcucci (1995) concluded in an experiment that phenolic acids including flavonoids derived from propolis and present in Manuka honey, exhibited weak antibacterial activity, have potentially contributing to the observed non-peroxide antibacterial activity (NPABA).

Isolation of a 5.8-kDa Component

Tonks, et. al. (2007) discovered a 5.8 kDa component in Manuka honey that stimulates human immune cells via TLR4, or the cytokine induction in human monocytes. This component was determined to be directly involved in the mechanism which stimulates innate immune cells to respond. While it is unknown whether this moiety is unique to Manuka honey or universally present in all honeys, it does appear to play a role in the antimicrobial activity observed in Manuka honey (Tonks, et. al., 2007).

Anti-Biofilm Activity in Manuka Honey in Combination with Antibiotics

Manuka honey has been proven to inhibit in-vitro antibiofilm experiments. More specifically, Gentamicin and Manuka honey produced an additive interaction against P. aeruginosa biofilms and a synergistic interaction with vancomycin against S. aureus biofilms (Campeau & Patel, 2014). When Manuka honey dressings were utilized in conjunction with oxacillin, tetracycline, imipenem, and mupirocim; a synergistic effect was achieved against MRSA (Carter, et, al., 2016). The findings suggested that the combined therapeutic intervention of Manuka honey wound dressings and antimicrobials could lower the dosage of antimicrobial medications required to inhibit the biofilm as well as prevent the development of resistance.

Manuka honey's bactericidal action is associated with non-peroxide antibacterial activity (NPABA). In-vitro isolates with multi-drug resistant phenotypes have not demonstrated any reduction in their susceptibility nor generated any honey-resistant strains to Manuka honey when administered at inhibitory levels under normal laboratory conditions (Carter, et, al., 2016). While bactericidal effects were seen in both planktonic cultures and biofilms, a higher concentration of Manuka honey was required to inhibit sessile bacteria, compared to its counterpart, free-living bacteria (Hammond, et. al., 2014). Test strains of well-known pathogenic bacteria were inhibited at lower concentrations in a liquid medium compared to on agar well diffusion plates (Hammond, et. al., 2014). This finding may be attributed to the ability of the components of Manuka honey to diffuse more readily and uniformly in a liquid medium due to the osmotic effects.

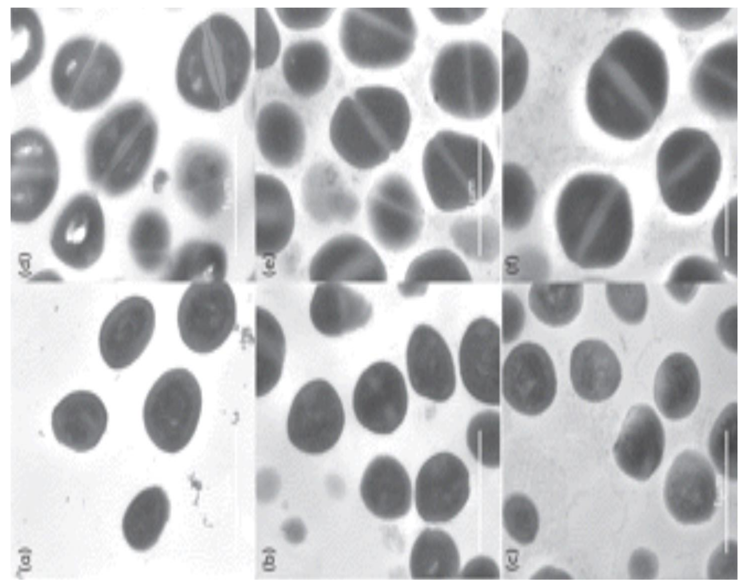


Figure 3: Inhibitory Effects of Manuka Honey on MRSA at x32000 magnification
(Jenkins, et. al., 2011)

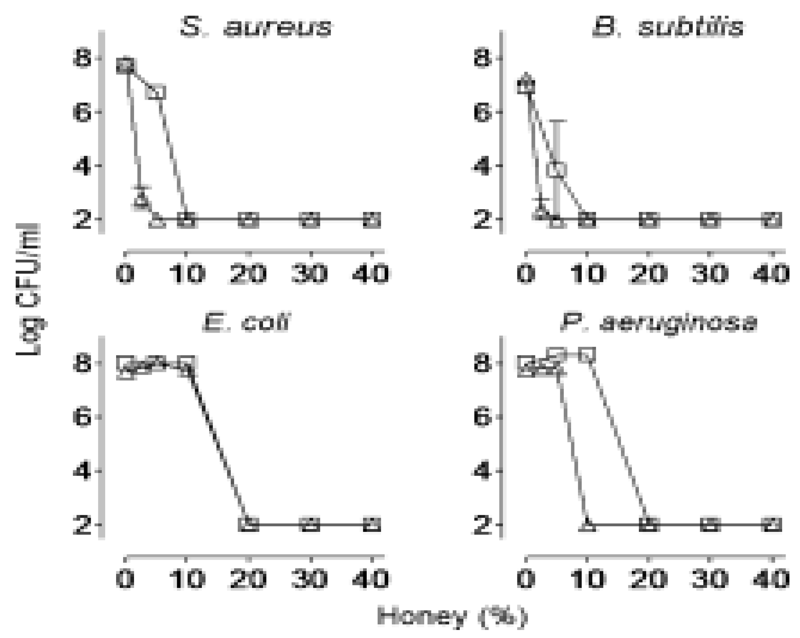


Figure 4: Antibacterial Activity of Manuka Honey: The Surviving Bacteria after Manuka Honey Was Administered
(Stewart & Costerton, 2001)

Further Applications

Gastrointestinal Ulcers

Almasaudi et. al. (2016) has performed experiments to assess gastroprotective effects associated with Manuka honey against ethanol-induced gastric ulcers in rats. After experimentation, the researchers were able to conclude that Manuka honey substantially lowered the rats' ulcer index, indicating that it protected the gastric mucosa from lesions and preserved gastric mucosal glycoproteins. Further research and experimentation is still needed in order to determine if Manuka honey is able to produce similar anti-ulcer outcomes in humans.

Anti-Cancerous Properties

Vallianou, Evangelopoulos, Skourtis, and Kazazi (2017) have confirmed with in-vitro experimentation, the anti-cancerous properties of Manuka honey via apoptosis. Scientists believe that Manuka honey induces caspase-9, which in turn, activates the caspase-3 executor proteins. These caspase enzymes are directly involved in the death receptor signaling pathway, which ultimately induces apoptosis (Vallianou, et. al., 2017). This contributes to the immune-protective and immune-modulatory activity, which is closely associated with anti-cancerous properties. In vivo testing has yet to be conducted. Therefore, Manuka honey's interaction in the human body is unknown and may inhibit or alter some of the anti-cancerous properties observed.

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