



**Prize Winner**

# Scientific Inquiry

## Year 7-8

### Kirra Dixon

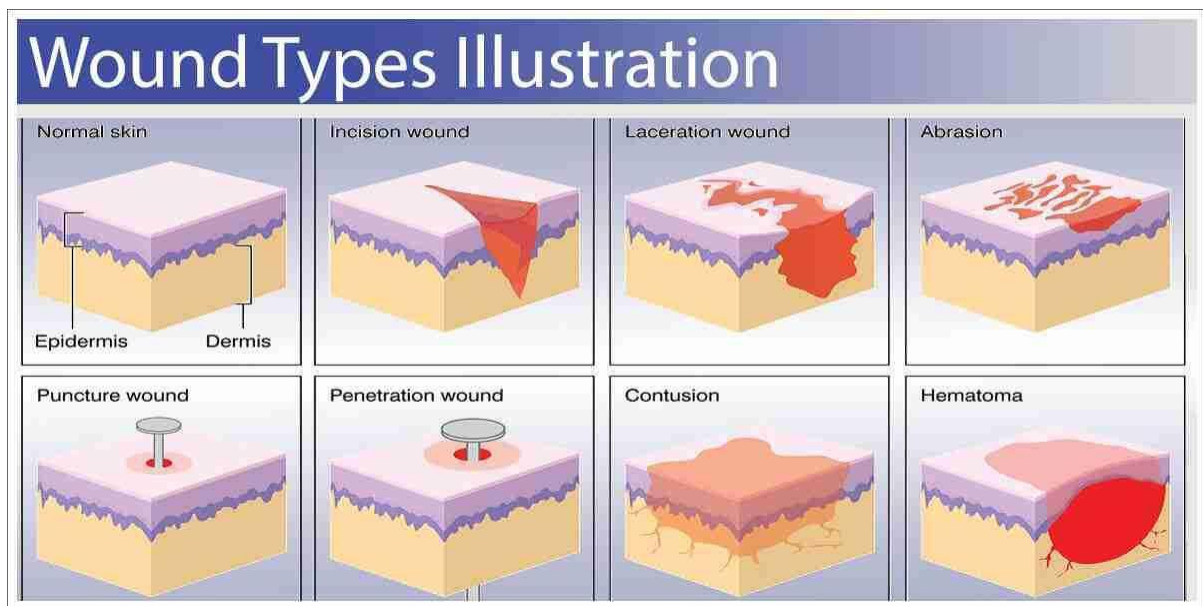
### Mercedes College



# Exploring the Effectiveness of Honey as a Natural Antiseptic: An Important Research Endeavour

## Introduction:

Caring for skin wounds is important to help them heal and prevent infection. Wounds can be grouped into two main types based on their depth and the layers of tissue they affect. The two main types of skin wounds are superficial or deep. Superficial wounds only affect the outer layer of the skin, known as the epidermis. Types of superficial wounds include abrasions and scratches (*Figure 1*). Deep wounds go below the superficial layers of the skin and can involve underlying tissues such as the fat and muscle layers. Deep wounds include incisions, lacerations, tears, punctures and penetrations (*Figure 1*). All skin wounds and burns can easily get infected by bacteria, making it harder for the wounds to heal and can cause other health problems (Percival et al., 2010).



*Figure 1: The different types of skin wounds (Source: ResearchGate, n.d.).*

Antiseptics are substances that help clean wounds and kill bacteria, preventing infections and helping wounds heal faster (DermNet New Zealand, n.d.). Some common antiseptics are hydrogen peroxide, iodine, and alcohol-based solutions, often used to treat wounds (Leaper et al., 2013). Even though these antiseptics effectively kill bacteria, they can cause burns or allergic reactions. Consequently, offering consumers an effective but natural alternative remedy such as honey is important.



*Figure 2: Allergic Reaction from using an antiseptic (Source: Medical News Today, n.d.).*

This experiment is important in the area of wound care. Its overall goal is to investigate the potential of honey as a natural antiseptic for treating skin wounds. The objectives of this experiment are to:

1. Find out if honey is as effective at preventing bacterial growth as the commonly used antiseptic, Dettol.
2. To determine the best type of honey (manuka, wildflower or clover) for stopping bacterial growth.

Honey has been recognised for its antibacterial properties for many years. It has a high sugar content, low pH (has an acidic property between 3.2-4.5) and contains hydrogen peroxide, which makes it good at fighting bacteria (Molan, 2001). Manuka honey, which comes from New Zealand, is especially known for its strong antibacterial effects because it has a unique substance called methylglyoxal (MGO) (Adams et al., 2008). Wildflower and clover honey are also known to have antibacterial properties.

Using honey as a natural antiseptic has many benefits. Honey helps keep the wound moist, which is good for healing and can reduce scarring (Subrahmanyam, 2007). It also has anti-inflammatory properties, which means it can reduce pain and swelling. Plus, honey is natural and usually does not cause bad reactions like some chemical antiseptics can (Molan, 2006).

If honey's antimicrobial properties can reduce our use of antibiotics, resistant bacterial strains might not develop so quickly (Kwakman et al., 2010). If this experiment can determine which type of honey is the most effective at combating infections, honey can then be seen as an accessible and affordable treatment option, particularly in areas that have limited access to antibiotics (Mandal & Mandal, 2011)

This experiment compares the effectiveness of honey as a natural antiseptic to a chemically made antiseptic, Dettol. It will also help us understand which honey might be the best choice for treating wounds naturally. To achieve this, sterile forceps will be used to create wells in agar plates for honey samples, while three plates will serve as controls. E. coli bacteria will be spread on the agar surfaces, and 0.5mL of different honey samples, along with a Dettol solution, will be added to the wells. Each plate will be labelled, sealed with parafilm, and incubated at 37°C for 48 hours. After incubation, zones of inhibition around the wells will be photographed and measured with a ruler. The diameters of these zones will be used to calculate the effectiveness of each sample in inhibiting bacterial growth, providing both qualitative and quantitative results.

**Aim:**

1. To find out if honey is as effective at preventing bacterial growth as the antiseptic Dettol.
2. To determine if honey prevents bacteria growth and, if so, which type of honey prevents bacteria growth the best.

**Hypothesis:**

1. I predict that Dettol will be better at preventing bacterial growth compared to honey.
2. I predict that Manuka honey will prevent bacteria growth the best of all of the honey samples.

**Variables:**

**Independent variable:** The different samples of honey (Manuka, Wildflower and Clover) and the Dettol solution.

**Dependent variable:** The size of the area around the honey well or Dettol well where bacteria did not grow (zones of inhibition).

**Controlled variables:**

1. All nutrient agar plates (both the control plate and plates containing honey samples) need to be exposed to a constant temperature (37°C) to make sure that bacteria growth conditions are the same.
2. Type of bacteria used – Escherichia coli
3. Incubation time (48 hours)
4. Using the same batch of agar plates
5. The same measurement technique (using a ruler) will be used to measure the inhibition zones around the honey wells.

**Materials:**

1. X15 nutrient agar plates
2. Different types of honey samples (Manuka, Wildflower and Clover honey).
3. X1 bottle of Dettol solution
4. X1 sterile spreader
5. X1 incubator (set to 37°C)
6. Bacterial culture (Escherichia coli).
7. X1 sterile forceps
8. X1 Sharpie marker for labelling the agar plates
9. Parafilm for sealing the agar plates
10. X4 teat pipettes.

**Method:**

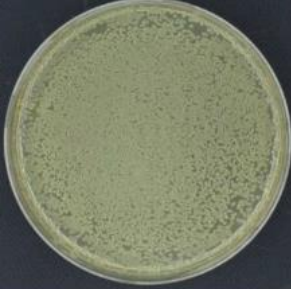
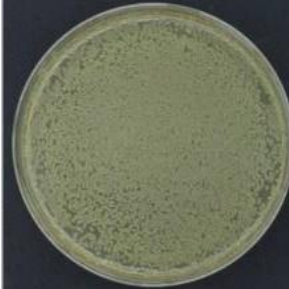

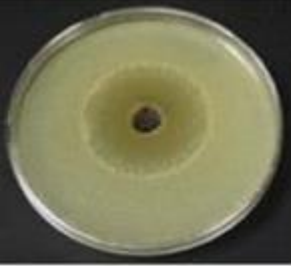
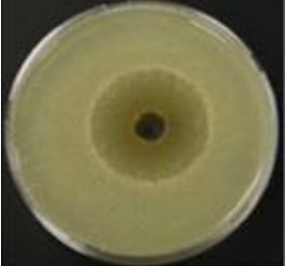


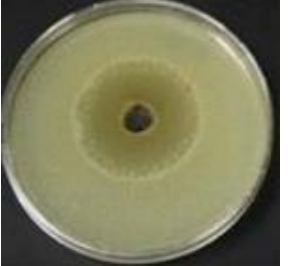

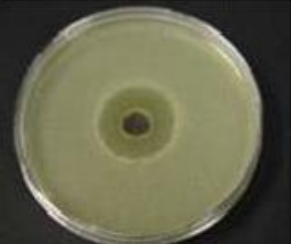

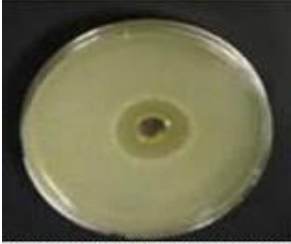
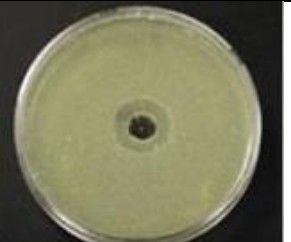

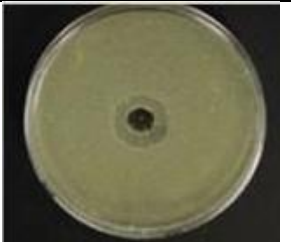
1. Using sterile forceps, one well was created in each agar plate where the honey samples were placed, except for the three plates, which served as a control (no honey).
2. The bacterial culture (E. coli) was spread onto the surface of the agar plates using a sterile spreader.
3. A teat pipette was used to add 0.5mL of one type of honey sample to the well of one agar plate. For the same honey sample, the process was repeated twice.
4. Step 3 was repeated for the other honey samples.
5. Step 3 was repeated for the Dettol solution.
6. Each agar plate was labelled with a Sharpie marker.
7. The Petri dishes were sealed with parafilm to stop contamination.
8. The agar plates were incubated at the appropriate temperature for human bacterial growth (37°C) for 48 hours.
9. After the incubation period, the agar plates were observed for zones of inhibition around the honey wells. Zones of inhibition were clear areas where bacterial growth was inhibited.
10. To show qualitative results, photos were taken of each petri dish and carefully placed on a table.

11. For quantitative results, the diameter of the zones of inhibition was measured for each sample using a ruler. This provided a numerical measure of each honey sample and the Dettol sample's effectiveness at stopping bacterial growth. An average diameter for each sample was calculated, along with the area of bacterial growth inhibition and the percentage of bacterial growth inhibition.

**Results:**

**1. Qualitative Results**

**Table 1: The Effectiveness of Different Types of Honey in Stopping Bacterial Growth**

Sample	Area of the Zone of Bacterial Inhibition (mm <sup>2</sup> )		
	Trial 1	Trial 2	Trial 3
Control			
Dettol			
Manuka honey			
Wildflower honey			
Clover			

## 2. Quantitative Results

**Table 2: The Effectiveness of Different Types of Honey in Stopping Bacterial Growth**

Sample	Zone of Inhibition					
	Trial 1 Diameter (mm)	Trial 1 Diameter (mm)	Trial 1 Diameter (mm)	Average Diameter (mm)	Area of Bacteria Growth Inhibition (mm <sup>2</sup> )	Percentage of Bacteria Growth Inhibition (%)
Control	0	0	0	0	0	0
Dettol	55.9	56.1	56.1	$\frac{55.9+56.1+56.1}{3}$ =56.0	$A = \pi r^2$ $= 3.14 \times 28^2$ =2461.8	$\frac{2461.8}{7853.9} \times 100$ =31%
Manuka	51.7	51.8	51.7	$\frac{51.7+51.8+51.7}{3}$ 51.8	$A = \pi r^2$ $= 3.14 \times 25.9^2$ =2106.3	$\frac{2106.3}{7853.9} \times 100$ =27%
Wildflower	33.3	33.1	33.1	$\frac{33.3+33.1+33.1}{3}$ =33.2	$A = \pi r^2$ $= 3.14 \times 16.6^2$ =865.3	$\frac{865.3}{7853.9} \times 100$ =11%
Clover	14.2	14.1	14.2	$\frac{14.2+14.1+14.1}{3}$ =14.2	$A = \pi r^2$ $= 3.14 \times 7.1^2$ =158.2	$\frac{158.2}{7853.9} \times 100$ =2%

Note: The area of a petri dish is  $A = \pi r^2$ , where  $r = 50\text{mm}$  (the radius of a petri dish)

$$= 3.14 \times 50^2$$

$$= 7853.9$$

### Discussion:

The experiment investigated the antimicrobial properties of various types of honey: Manuka, wildflower, and clover, compared to a standard antiseptic (Dettol) and a control.

The percentage of bacteria growth inhibition for each sample was as follows: Dettol at 31%, Manuka honey at 27%, wildflower honey at 11%, clover honey at 2%, and the control at 0%.

The results indicate a clear trend in the effectiveness of the antibacterial activity of the tested substances. Dettol demonstrated the highest bacterial growth inhibition at 31%, followed closely by Manuka honey at 27%. Wildflower honey showed moderate inhibition at 11%, while clover honey demonstrated minimal inhibition at 2%. As expected, the control showed no inhibition, establishing a baseline for comparison. Therefore, my results support both hypotheses, which were:

1. Dettol will be better at preventing bacterial growth compared to honey.
2. Out of all of the honey samples, Manuka honey will prevent bacteria growth the best.

The control sample, which had 0% inhibition, served as a baseline to determine the natural growth rate of bacteria without any antimicrobial substances. This is important for accurately assessing the effectiveness of the other substances tested. By comparing the treated samples against the control, the experiment can isolate the impact of each substance on bacterial growth inhibition.

The properties of each substance can explain the varying levels of bacterial growth inhibition observed. Dettol, a synthetic antiseptic, is specifically made with a chemical called Chloroxylonol to kill a broad spectrum of bacteria, which explains why it is the most effective substance at inhibiting bacterial growth (*Table 1 and Table 2*). Chloroxylonol disrupts the bacteria's cell membrane, causing leakage of many essential ions and other metabolites. It also denatures (unfolds) proteins found inside bacteria cells, including enzymes, which are essential for carrying out many vital reactions (Kramer et al., 2004).

Honey's low pH (between 3.2 and 4.5) and high sugar content create a hyperosmotic environment that inhibits bacterial growth. The low pH disrupts bacterial cell functions, while the high osmolarity draws water out of bacterial cells because water travels from an area of high water concentration (inside the bacteria) to an area of low concentration (the honey environment) by osmosis. This leads to dehydration and, ultimately, cell death of the bacterial cells, which explains why all honey samples showed some bacteria growth inhibition (*Table 1, Table 2*) (Molan, 2001).

Manuka honey is known for its unique antimicrobial properties, largely because it has a chemical called methylglyoxal (MGO), which contributes to its antibacterial effectiveness. MGO disrupts the bacterial cell membrane and damages the bacteria's DNA, affecting its ability to divide by binary fission (Kwakman et al., 2010). MGO also alters the structure and function of proteins inside bacteria, causing enzymes to stop carrying out important cellular processes (Kwakman et al., 2010). Although less potent than Manuka, wildflower honey contains various phytochemicals that provide some antibacterial activity (Kwakman et al., 2010). Clover honey, however, has lower levels of these active compounds, resulting in minimal inhibition (Kwakman et al., 2010).

Random errors are shown as the natural variation of the diameter for each trial for a given sample. The natural variation in the results is called imprecision. One potential random error would be the amount of Dettol or honey added to each well. The measured amount of Dettol or honey was 0.5mL, but slight variations could have occurred. If one well had a slightly larger amount than another well, then this might cause greater antibacterial activity. Another random error was the number of bacteria spread onto the agar in each Petri dish. Some plates might randomly have more bacteria than others. The Petri dishes with more bacteria probably showed less antibacterial activity because more bacterial cells had to be killed. Random errors cannot be eliminated, but their effects can be minimised. Therefore, to minimise the effects of these random errors, the number of trials would need to be increased, the experiment would need to be repeated a few more times and average diameter results calculated for each sample. Repeating the experiment and increasing the number of trials will also allow me to see if my results are reliable (getting similar results for each trial) for each substance.

Systematic errors are caused by human mistakes or errors in the method and affect the results' accuracy. One mistake was that the area of bacteria growth inhibition was not exactly circular. Therefore, I decided to measure the largest distance (diameter) across the zone of inhibition, which would have affected the accuracy of the results. The percentage of bacteria growth inhibition would have been larger than if I had decided to measure the shortest distance (diameter) across the zone of inhibition. Another systematic error could be cross-contamination, which could have occurred between the samples if a clean pipette was not used. Suppose the pipette previously used to pipette Dettol into a well was used for a honey sample. In that case, the honey sample may have shown greater antibacterial activity than it should.

To better understand honey as an effective natural antiseptic, systematic errors must be considered when repeating the practical to make sure that the results are valid and reliable.

## Conclusion:

In this experiment, I aimed to test the hypotheses that:

1. Dettol will be better at preventing bacterial growth compared to honey.
2. Manuka honey was expected to show the highest bacterial inhibition growth among the honey samples.

The results supported my hypothesis. Dettol, a chemical disinfectant, showed the highest bacterial growth inhibition at 31%. Manuka honey was the most effective among the honey samples, with 27% inhibition, followed by wildflower honey at 11% and clover honey at 2%. The control, which had no treatment, showed 0% inhibition.

These findings confirm that Dettol was better at preventing bacterial growth than honey, most likely because it contained a synthetic chemical called chloroxylenol, which disrupts bacterial cell membranes. The results also showed that Manuka honey, with its high levels of methylglyoxal (MGO), had the strongest antibacterial properties out of all honey samples. MGO inhibits bacterial growth by disrupting their cell membrane and damaging the bacteria's DNA, affecting its ability to divide by binary fission. Wildflower honey has some antibacterial effects due to its natural phytochemicals, although it is not as potent as Manuka honey. Clover honey, with fewer antibacterial compounds, showed the least inhibition of bacterial growth.

Overall, the experiment demonstrated that different types of honey have varying abilities to inhibit bacterial growth, with Manuka honey being the most effective among the tested kinds of honey. The data supported our hypothesis, showing that natural substances like honey can have significant antimicrobial properties, which vary depending on their composition. This experiment highlights the potential of using honey, particularly Manuka honey, as a natural antibacterial agent.

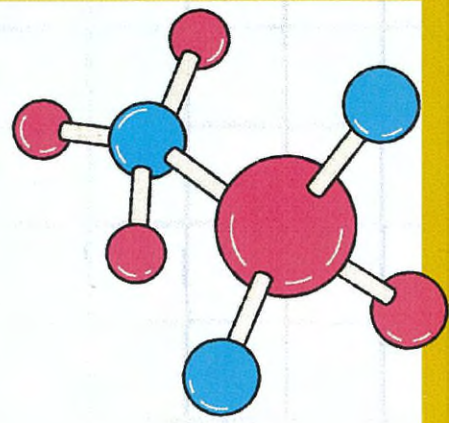
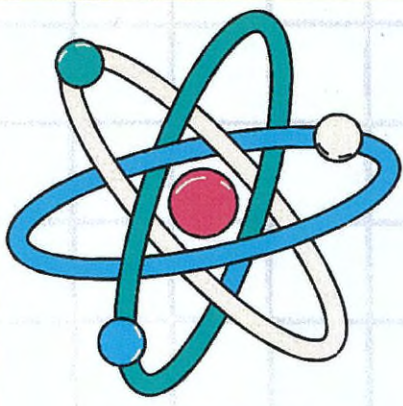
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6. Molan, P. C. (2001). Potential of honey in the treatment of wounds and burns. *American Journal of Clinical Dermatology*, 2(1), 13-19.
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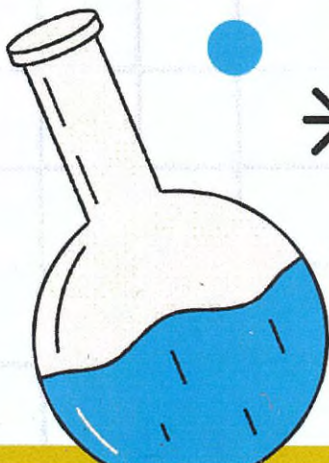
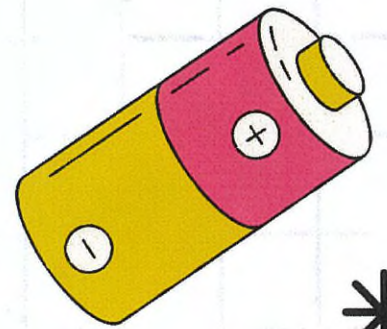


8. Percival, S. L., Bowler, P. G., & Russell, D. (2010). Bacterial resistance to silver in wound care. *Journal of Hospital Infection*, *60*(1), 1-7.
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Word count: 1893



# SCIENCE INQUIRY



KIRRA DIXON

date: 9/4/24

## introduction

name: kirra Dixon

category: Scientific Inquiry

School: Mercedes college

OSA coordinator: caroline Beekman

## The Scientific Method

The scientific method is a process that scientists use to discover observations and obtain answers. Most of the time, scientists use this method, but not all scientists have to follow it. It's a way of doing research to find any unknown problems, collecting information, and a way to form a hypothesis and investigate.

# The Scientific Method

## Steps to the Scientific Method:

### ① Purpose/question

The journey of all science experience begins when someone starts to wonder why things happen in nature the way they do. And that journey always starts with asking a question.

### ② Research

Background research is a key step in any science prac. keeping track of your sources is crucial for accurately mentioning references. Better understanding makes experiments easier to conduct. Therefore, keeping track of where you get your information helps make your research strong.

### ③ Hypothesis

The third step is to propose a hypothesis. This is essentially a well educated prediction stated as a statement, which helps predict the result of an experiment. While typically focusing on cause and effect, hypotheses are made to explore connections between variables.

IF \_\_\_\_\_, Then \_\_\_\_\_, Because \_\_\_\_\_.

# The Scientific Method

## Steps continued:

### ④ Experiment

Next, an experiment is designed and performed. The experiment must have an independent and dependent variable. Furthermore, conducting the experiment at least once or twice is vitally important to support the reliability of the gathered data.

### ⑤ Data/Analysis

The fifth step involves recording observations and search into the significance of the gathered data. This data is commonly organized in tables or graphs for simplicity. If the experimental outcomes go different ways from the anticipated results outlined by the hypothesis, it indicates a disapproval of the theory.

### ⑥ Conclusion

In the final phase, you determine whether to keep or disprove your hypothesis. Understandably, there exists no absolute right or wrong in an experiment; each result holds its own effectiveness. At this point, it's really important to share your findings in a way that others can understand easily.

## Sources accessed:

• <https://www.sciencebuddies.org/science-fair-projects/science-fair/steps-of-the-scientific-method>

Date: 10/4/24

◦ <https://www.thoughtco.com/steps-of-the-scientific-method-p2-60645>

## The Project

### Exploring the Effectiveness of Honey as a Natural Antiseptic

I am interested in investigating whether honey is just as effective as an antimicrobial agent compared with everyday skin antiseptics. Additionally, I aim to identify which type of honey demonstrates the highest effectiveness in combating microbial activity. This research could provide valuable insights into the potential use of honey as a natural alternative to traditional antiseptics in wound care and infection management. Everyday skin antiseptics contain chemicals like iodine, chlorhexidine and alcohol that can cause skin irritation or damage in some people, especially if used in high concentrations or over long periods of time.

# Background Research

## 1. The 2 types of Skin Wounds

Wounds can be grouped into two main types based on their depth and the layers of tissue they affect. The two main types of wounds are superficial and deep.

### 1. Superficial Wounds:

Superficial wounds only affect the outer layer of the skin, known as the epidermis. These wounds usually involve a small amount of bleeding and generally heal quickly without a lot of scarring. Types of superficial wounds include:

- Abrasions: An abrasion occurs when the skin is scraped or rubbed against a rough surface, such as asphalt or gravel. It results in the removal of the superficial layers of the skin, causing pain and often leaving a raw, red area. [1]

- Scratches: A scratch is a shallow wound caused by a sharp object, such as fingernails or thorns, which lightly breaks the skin's surface. Scratches may be painful but usually heal quickly without any problems. [2]

- Superficial lacerations: A superficial laceration is a shallow cut that does not penetrate deeply into the skin layers. It may result from sharp objects like knives or glass and typically involves minimal bleeding. [3]

## Background Research



<https://www.cprfirstaidtraining.co.uk/advice-tips/minor-wounds-cuts-and-grazes/>

Reference:

### 2. Deep Wounds:

Deep wounds go below the superficial layers of the skin and can involve underlying tissues, such as the dermis (skin), fat, muscles, tendons, or bones. These wounds often need medical attention and may take longer to heal. Types of deep wounds include:

- Incision: An incision is a clean, straight cut caused by a sharp object, such as a knife or scalpel. It may penetrate deeply into the skin layers and can result in a lot of bleeding. Depending on the depth and location, an incision might need stitches. [4]

- Laceration: A laceration is a jagged or irregular wound caused by a tearing or crushing force. It can affect many layers of tissue and, therefore, cause extensive bleeding. Lacerations often happen due to accidents, falls, or blunt trauma. [5]



## Background Research

-Puncture: A Puncture wound is caused by a sharp, pointed object piercing the skin, such as nails, needles, or teeth. Puncture wounds can be deep and potentially damage internal organs if not treated straight away. Because puncture wounds have a narrow opening, they are at a higher risk of infection. [6]

-Tears: A tear happens when a portion of the skin and underlying tissue is forcibly torn away from the body. This type of injury can be serious and may require surgery to repair the damaged tissue. [7]

Deep wounds often need thorough cleaning, removal of dead tissue, and sometimes stitches to help with healing and prevent infection. Additionally, deep wounds may cause scarring or permanent injury, especially if they involve structures like nerves or tendons. Therefore, immediate medical treatment is really important for treating a person who has deep wounds.



Reference: 3  
<https://westcoastwound.com/how-to-help-open-wounds-heal-faster/>

# Background Research

## References:

1. KidsHealth from Nemours. (2023). Cuts, Scrapes, and Scar care. Retrieved from <https://kidshealth.org/en/kids/cut-scrape.html>
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3. Stanford children's health. (n.d). Wounds. Retrieved from <https://www.stanfordchildrens.org/en/topic/default?id=wounds-90-P02686>
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# Background Research

## Types of Antiseptics in Wound Care:

An antiseptic is a substance that stops the growth of bacteria and viruses on the skin and helps prevent infections (DermNet NZ, n.d.).

Antiseptics play a critical role in wound care by preventing or reducing the risk of infection, which is essential for healing. They are substances applied to living tissues to stop the growth of germs in and around the wound area (DermNet NZ, n.d.).

- 1. Preventing Infection:** Antiseptics prevent infection by killing or stopping the growth of microorganisms such as bacteria, fungi, and viruses. By reducing the number of germs present in and around the wound, antiseptics help minimise the risk of infection and the development of complications such as sepsis (DermNet NZ, n.d.).
- 2. Promoting Healing:** In addition to preventing infection, antiseptics also promote wound healing by creating a clean environment that allows the tissue to repair. By reducing the number of germs, antiseptics support the body's natural healing processes, causing faster wound closure (DermNet NZ, n.d.).

## Background Research

Commonly used antiseptics in wound care include:

- Iodine-based antiseptics: can kill a wide range of bacteria and viruses (DermNet NZ, n.d.).
- Chlorhexidine: effective against a wide range of bacteria and is commonly used as a skin disinfectant (DermNet NZ, n.d.).
- Alcohol: Ethanol and isopropyl alcohol are commonly used as disinfectants for skin wounds (DermNet NZ, n.d.).
- Hydrogen peroxide: Although debated because it can potentially damage healthy tissue, hydrogen peroxide releases oxygen, which helps kill bacteria and promote healing (DermNet NZ, n.d.).
- Silver-based antiseptics: Silver has strong antimicrobial properties and is used to prevent infection in wounds (DermNet NZ, n.d.).

It's important to use antiseptics carefully and to consider factors such as the type and severity of the wound and the risk of allergic reactions (DermNet NZ, n.d.).

### References:

DermNet New Zealand. (n.d). Antiseptic. Retrieved from <https://dermnetnz.org/topics/antiseptic>

# Background Research

Different antimicrobial properties of honey samples has several benefits to society

1. Improved Wound Healing Treatments: Understanding which types of honey have the strongest antimicrobial properties can lead to the development of more effective wound-healing treatments. Certain types of honey, such as Manuka honey, have been found to have strong antibacterial activity against a wide range of bacteria, including antibiotic-resistant strains like MRSA. (Methicillin-resistant *Staphylococcus aureus*) (Irish et al., 2011; Copper et al., 2001).
2. Reduced Antibiotic Resistance: Honey offers a natural solution for combating antibiotic-resistant bacteria. Its antimicrobial properties can reduce reliance on antibiotics, thereby slowing the emergence of resistance strains. (Kwakman et al., 2010).
3. Accessible and Affordable Healthcare: If certain types of honey are found to be effective in combating infections, they could serve as accessible and affordable treatment options, particularly in resources-limited settings where access to conventional antibiotics may be limited (Mandal & Mandal, 2011).
4. Potential for Home Remedies: With education, people can use honey as a natural remedy for minor wounds and infections at home (Alvarez Suarez et al., 2014).

## Background: Research

Different antimicrobial properties of honey samples has several benefits to society - continued:

### 5. Support for Beekeeping and Agriculture:

Investigating honey's antimicrobial properties can benefit beekeeping and agriculture by improving practices to maximise the process of high-quality honey that has strong antimicrobial properties (Willix et al., 1992).

### 6. Research and Innovation: Continued research into honey's antimicrobial properties can further develop new solutions for treating infections (Irish et al., 2011; Willix et al., 1992).

## Reference List:

- Alvarez-Suarez JM, Gasparri M, Forbes-Hernandez TY, Mazzoni L, Giampieri F. The composition and Biological Activity of Honey: A Focus on Manuka Honey. *Foods*. 2014; 3(3): 420-432. doi:10.3390/foods3030420.
- Cooper BA, Molan PC, Krishnamoorthy L, Harding KG. Manuka honey used to heal a recalcitrant surgical wound. *European Journal of Clinical Microbiology & Infectious Diseases*. 2001; 20 (10): 758-759. doi:10.1007/s100960100585.

# Background Research

## Reference List:

- Irish J, Blair S, Carter DA. The Antibacterial Activity of Honey Derived from Australian Flora. PLoS One. 2011; 6(3). doi:10.1371/journal.pone.0018229.
- Kwakman PH, te Velde AA, de Boer L, et al. How honey kills bacteria. FASEB Journal. 2010; 24(7):2576-2582. doi:10.1096/fj.09-150789.
- Mandal MD, Mandal S. Honey: its medicinal property and antibacterial activity. Asian Pacific Journal of Tropical Biomedicine. 2011; 1(2):154-160. doi:10.1016/S2221-1691(11)60016-6.
- Willix DJ, Molan PC, Horfoot CG. A comparison of the sensitivity of wound-infecting species of bacteria to the antibacterial activity of manuka honey and other honey. Journal of Applied Bacteriology. 1992; 73(5):388-394. doi:10.1111/j.1365-2672.1992.tb04919.x.

These references support the claims about honey's antimicrobial properties and their potential applications in wound healing, healthcare, agriculture, and research.

13/6/24

# Practical Design

Exploring the Effectiveness of Honey as a Natural Antiseptic

Aim:

1. To find out if honey is as effective at preventing bacterial growth as the antiseptic Dettol.
2. To determine if honey prevents bacteria growth and, if so, which type of honey prevents bacteria growth the best.

Hypothesis:

1. I predict that Dettol will be better at preventing bacterial growth compared to honey.
2. I predict that out of all the honey samples Manuka honey will prevent bacteria growth the best.

Variables:

Independent Variable: The different samples of honey (Manuka, Wildflower and Clover) and the Dettol solution.

Dependant variable: The size of the area around the honey well or Dettol well where bacteria didn't grow



13/6/24

# Practical Design

Exploring the Effectiveness of Honey as a Natural Antiseptic

## Controlled Variables:

1. All nutrient agar plates (both the control plate and plates containing honey samples) need to be exposed to a constant temperature ( $37^{\circ}\text{C}$ ) to make sure that bacteria growth conditions are the same.
2. Type of bacteria used - *Escherichia coli*
3. Incubation time (48 hours)
4. Using the same batch of agar plates.
5. The same measurement technique (using a ruler) will be used to measure the inhibition zones around the honey wells.

## Materials:

1. X5 Nutrient agar plates
2. Different types of honey samples (e.g., Manuka, wildflower, and clover honey)
3. Dettol solution
4. X1 Sterile spreader
5. An incubator (set to  $37^{\circ}\text{C}$ )
6. Bacterial culture (*Escherichia coli*)
7. X1 Sterile forceps
8. X1 Sharpie marker for labelling.
9. Parafilm for sealing the agar plates.
10. X4 Test pipettes

13/6/24

# Practical Design

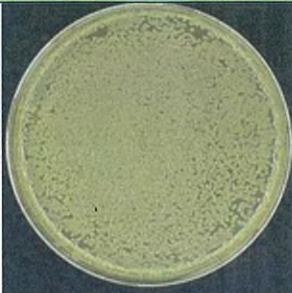
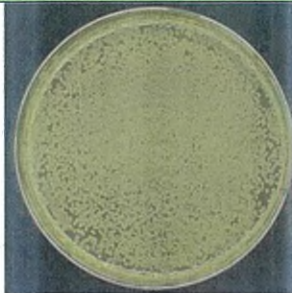
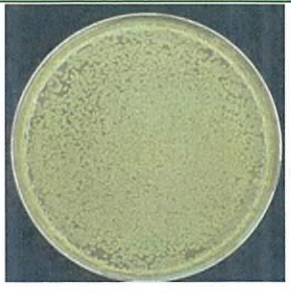
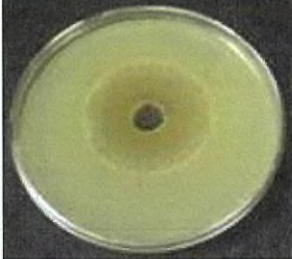

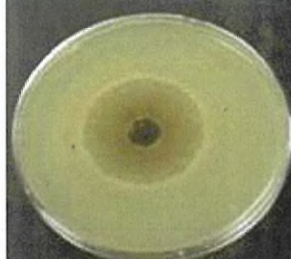

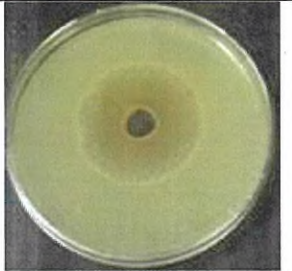
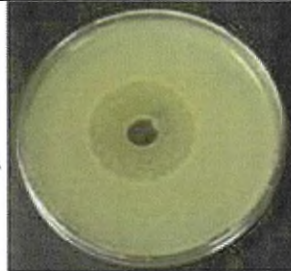
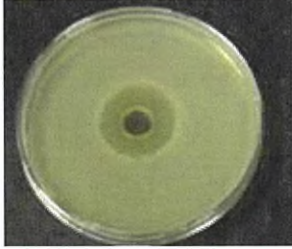
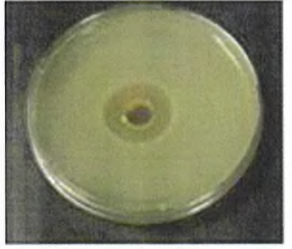
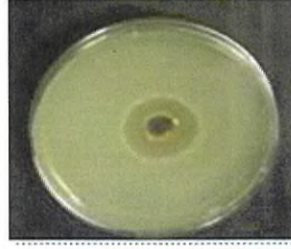
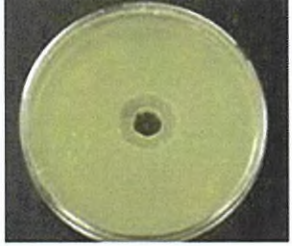

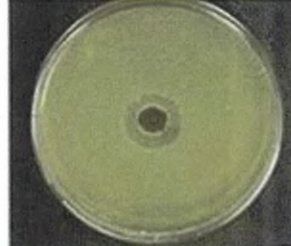
Exploring the Effectiveness of Honey as Natural Antiseptic

Method:

1. Using sterile forceps, create one well in each agar plate where the honey samples will be placed.
2. Use a sterile spreader to spread the bacterial culture (E. coli) onto the surface of the agar plates.
3. Using a test pipette, add 0.5 mL of one type honey sample into the well of one agar plate. Repeat the process twice more for the same honey sample. Repeat this process for all honey samples.
4. Repeat step 3 for Dettol solution.
5. Repeat step 3 for the control (no honey).
6. Label each agar plate.
7. Seal the Petri dishes with parafilm to stop contamination.
8. Incubate the agar plate at the appropriate temperature for bacterial growth in humans (usually around  $37^{\circ}\text{C}$ ) for 48 hours.
9. After the incubation period, observe the agar plates for zones of inhibition around the honey wells. Zones of inhibition are clear areas where bacterial growth is inhibited.
10. Measure the diameter of the zones of inhibition using a ruler. This provides a quantitative (numerical) measure of each honey sample and the dettol sample effectiveness at stopping bacterial growth.

14/6/24

Results (Qualitative)

Sample	Area of the Zone of Bacterial Inhibition (mm <sup>2</sup> )		
	Trial 1	Trial 2	Trial 3
Control			
Dettol			
Manuka			
Wildflower			
Clover			

14/6/24

## Results (Quantitative)

The diameter of each zone of inhibition was measured with a ruler (mm), and the area of the zone of inhibition was calculated using the formula.

$$A = \pi r^2 \quad (\text{where } \pi = 3.14)$$

The zone of inhibition as a percentage has also worked out:

$$\frac{\text{Area of zone of inhibition}}{\text{Area of entire petri dish}} \times 100$$

### The Effectiveness of Different Types of Honey in Stopping Bacterial Growth

Sample	Zone of Inhibition					
	Trial 1 Diameter (mm)	Trial 1 Diameter (mm)	Trial 1 Diameter (mm)	Average Diameter (mm)	Area of Bacteria Growth Inhibition (mm <sup>2</sup> )	Percentage of Bacteria Growth Inhibition (%)
Control	0	0	0	0	0	0
Dettol	55.9	56.1	56.1	$\frac{55.9+56.1+56.1}{3}$ =56.0	$A = \pi r^2$ $= 3.14 \times 28^2$ =2461.8	$\frac{2461.8}{7853.9} \times 100$ =31%
Manuka	51.7	51.8	51.7	$\frac{51.7+51.8+51.7}{3}$ 51.8	$A = \pi r^2$ $= 3.14 \times 25.9^2$ =2106.3	$\frac{2106.3}{7853.9} \times 100$ =27%
Wildflower	33.3	33.1	33.1	$\frac{33.3+33.1+33.1}{3}$ =33.2	$A = \pi r^2$ $= 3.14 \times 16.6^2$ =865.3	$\frac{865.3}{7853.9} \times 100$ =11%
Clover	14.2	14.1	14.2	$\frac{14.2+14.1+14.1}{3}$ =14.2	$A = \pi r^2$ $= 3.14 \times 7.1^2$ =158.2	$\frac{158.2}{7853.9} \times 100$ =2%

Note: The area of a petri dish is  $A = \pi r^2$ , where  $r = 50\text{mm}$  (the radius of a petri dish)

$$= 3.14 \times 50^2$$

$$= 7853.9$$

15/6/24

## Analysis:

The Results showed:

**Control:** Bacteria grew to the point where nearly the entire petri dish was covered in E. Coli.

**Dettol:** Had the largest zone of bacterial inhibition.

**Honey Samples:** Manuka honey had the second largest zone of bacterial inhibition and Clover honey had the smallest zone.

Conclusion: ① Honey is effective at stopping bacterial growth, but it isn't as good as Dettol.

② Manuka honey is the most effective out of all the honey samples to stop bacterial growth and Clover honey was the worst at stopping bacterial growth.

# OSA RISK ASSESSMENT FORM

for all entries in  Models & Inventions and  Scientific Inquiry

This must be included with your report, log book or entry. One form per entry.

STUDENT(S) NAME: Kirra Dixon

ID: 0370-009

SCHOOL: Mercedes College

**Activity:** Give a brief outline of what you are planning to do.

My experiment is to investigate the potential of honey as a natural antiseptic for treating skin wounds. The objectives of this experiment are to:

1. Find out if honey is as effective at preventing bacterial growth as the commonly used antiseptic, Dettol.
2. To determine the best type of honey (manuka, wildflower or clover) for stopping bacterial growth.

In this experiment, sterile forceps will be used to create wells in agar plates for honey samples, while three plates will serve as controls. E. coli bacteria will be spread on the agar surfaces, and 0.5mL of different honey samples, along with a Dettol solution, will be added to the wells. Each plate will be labelled, sealed with parafilm, and incubated at 37°C for 48 hours. After incubation, zones of inhibition around the wells will be photographed and measured with a ruler. The diameters of these zones will be used to calculate the effectiveness of each sample in inhibiting bacterial growth, providing both qualitative and quantitative results.

**Are there possible risks? Consider the following:**

- Chemical risks: Are you using chemicals? ]
- Biological risks: Are you working with micro-organisms such as mould and bacteria?
- Sharps risks: Are you cutting things, and is there a risk of injury from sharp objects?
- Electrical risks: Are you using mains (240 volt) electricity? How will you make sure that this is safe? Could you use a battery instead?

**Also, if you are using other people as subjects in an investigation you must get them to sign a note consenting to be part of your experiment.**

Risks	How I will control/manage the risk
Metal forceps can accidentally be poked or poked in the eye	I will be careful when handling the metal forceps.
An incubator can cause an electrical shock if it is not wired properly.	Speak with the lab technician to find out if the incubator has been recently tested and tagged to make sure that it is safe.
E. coli bacteria culture can cause infection and illness when used with a spreader to spread bacteria on nutrient agar plates.	To prevent infection and illness, I will wear gloves, a lab coat and lab glasses when spreading bacteria on an agar plate. A teacher will also be guiding and supervising me to make sure that I am handling the bacteria culture with care.
Honey, Dettol and marker pen (active ingredient chloroxylonol) can cause allergic reactions.	To prevent a potential allergic reaction, I will wear gloves, a lab coat, and lab glasses. I will not eat the honey or sniff the marker pen, which would contain toxic chemicals and fumes.

(Attach another sheet if needed.)

**Risk Assessment indicates that this activity can be safely carried out**

RISK ASSESSMENT COMPLETED BY (student name(s)): Kirra Dixon

SIGNATURE(S): Kirra Dixon

By ticking this box, I/we state that my/our project adheres to the listed criteria for this Category.

TEACHER'S NAME: Caroline Beekman

SIGNATURE: [Signature] DATE: 2/6/24