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Potential Bioactivity of Carrot (*Daucus carota* L.) as a Health Protector Through Antioxidant, Antibacterial, and Antifungal Activities

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Abstract: Carrots are one of the most popular plants in Indonesia. In addition to being a plant with a flavor favored, carrots also contain various bioactivities that can be utilized for health. This study aims to investigate the bioactivity of carrot extract, which includes antioxidant, antibacterial, and antifungal. The method used in this study is DPPH, which aims to determine antioxidant activity, and the disc diffusion method to determine antibacterial and antifungal activity. Carrot extract was obtained using the maceration method with a ratio of 1:10 using ethanol solvent 96% for the antioxidant test, while for the antibacterial and antifungal tests, acetone solvent was used. The extracted carrots yielded a yield of 10.1% (v/b). Based on the results of phytochemical screening, it is known that carrots contain flavonoids, saponins, tannins, and alkaloids, as well as functional groups indicating the presence of beta-carotene in carrots through FTIR. Carrots show weak antioxidant activity with an IC_{50} value of 125.944 ppm. The inhibitory activity of carrot extract during 24 hours of observation are concentration 40% (0.7 mm), 60% (0.77 mm) and 80% (0.85 mm). Meanwhile, in the antifungal test, carrot extract showed negative results at concentrations 60 and 80%, while at a concentration of 100% showed an inhibition zone of 0.835 mm.

Keywords: Antibacterial; Antifungal; Antioxidant and carrot

Introduction

Carrot (*Daucus carota* L.) is a type of vegetable with a sweet taste. Carrot belongs to the Apiaceae family and is cultivated worldwide (Senas & Islamy, 2023). This plant has high nutritional content and is known as the primary source of β-carotene, which serves as a precursor to vitamin A (Astuti et al., 2024; Mai et al., 2022; Workneh et al., 2011). In addition, carrots are rich in various other bioactive compounds, such as polyphenols, flavonoids, and essential oils, which contribute to their biological activities, including antioxidant, anti-inflammatory, anticancer (anticarcinogenic), antimicrobial, cardioprotective, and antidiabetic properties (through the inhibition of enzymes involved in carbohydrate and lipid metabolism (Ahmad et al., 2019; Maresca et al., 2024; Singh et al., 2021). Natural antioxidants derived from plant sources are beneficial for health (Fardani et al., 2025). These antioxidant compounds help neutralize free radicals, which are chemically reactive species with unpaired electrons that readily interact with proteins, lipids,

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carbohydrates, or DNA (Agustikawati et al., 2017; Lavlinesia et al., 2023; Lestari et al., 2023).

The antioxidant properties of carrots are closely linked to their β-carotene and phenolic compound content. These bioactive compounds are recognized for their nutraceutical effects and health benefits. Four types of phytochemicals found in carrots include phenolics, carotenoids, polyacetylenes, and ascorbic acid. These compounds are known for their antioxidant activity, which can neutralize free radicals and protect cells from oxidative damage associated with degenerative diseases such as cancer, cardiovascular diseases, and premature aging (Ahmad et al., 2019; Ibroham et al., 2022). Previous studies have demonstrated that carrot extracts possess significant potential as effective natural antioxidants (Azizuddin & Qadeer, 2017).

Besides being an antioxidant, carrots are also known for their antimicrobial activity. This is attributed to their secondary metabolite content, which includes phenolics, flavonoids, alkaloids, terpenoids, and carotenoids (Babic et al., 1994; Fartoosi et al., 2025; John et al., 2018; Søltoft et al., 2010). The antimicrobial activity in carrots includes antibacterial and antifungal properties. Several studies have revealed that bioactive components in carrots can inhibit the growth of both Gram-positive and Gram-negative bacteria, as well as various pathogenic fungi (Ratajczak et al., 2023; Shindia et al., 2024; Trigo et al., 2020; Vegara et al., 2011; Yuan et al., 2021).

The antibacterial and antifungal activities of carrots can be utilized in the development of natural-based products for microbial infection treatment and as a natural preservative in the food industry (Shindia et al., 2024). These findings not only highlight the potential health benefits of incorporating carrots into our diets but also suggest innovative avenues for research into sustainable alternatives for both healthcare and food preservation. As the demand for natural solutions continues to rise, further exploration of carrot-derived compounds could lead to significant advancements in both fields.

Several journals have examined the antimicrobial activity of carrots and their applications in various fields. A study by Sirait (2016) demonstrated that ethanol extracts from carrots exhibited inhibitory effects against *Staphylococcus aureus* and *Escherichia coli*. Another study by Sharma et al. (2012) revealed that the phenolic content in carrots has potent antifungal properties against *Candida albicans*. Evaluated the effects of combining carrot extracts with other bioactive compounds to enhance antibacterial efficacy against foodborne pathogens. *Escherichia coli* is a bacterium with dual characteristics: it can aid digestion but can also cause diarrhea if its population is unregulated. Therefore, its presence in the intestines must be controlled (Rasmi et al., 2023). Meanwhile, *Candida albicans* is an opportunistic fungus that can adversely affect the digestive system, mouth, skin, and reproductive organs (Winarni et al., 2023).

The bioactive compounds in carrots, such as antioxidants, antifungal, and antibacterial agents, can be extracted using maceration methods with polar solvents (Putra, 2022; Utomo et al., 2018; Yuningsih & Putra, 2025). Polar solvents suitable for extraction include ethanol, methanol, acetone, water, and acetic acid (Runtuboi et al., 2024).

The presence of these bioactivities plays a crucial role in increasing the appeal of carrot consumption. Therefore, this study aims to screen the bioactivities of carrots, including their antioxidant, antibacterial, and antifungal properties. Additionally, this research seeks to identify the active compounds responsible for these activities and evaluate their effectiveness against various pathogenic microorganisms. By gaining a more profound understanding of the bioactive potential of carrots, this study is expected to provide insights into their utilization not only as a nutritional source but also as a natural ingredient in the development of health products, functional foods, and herbal-based therapies.

Method

Preparation of Carrot Extract

The carrots used in this study were fresh, large-sized carrots. After being thoroughly washed, the carrots were thinly sliced and dried at room temperature for seven days to prevent the degradation of bioactive compounds due to exposure to high temperatures. Once dried, the carrots were ground into powder. The maceration process was then carried out using acetone as the solvent for antibacterial (*Escherichia coli*) and antifungal (*Candida albicans*) tests, while 96% ethanol was used for the antioxidant test.

The difference in solvent selection for antimicrobial and antioxidant tests was based on the polarity of the respective compounds. Acetone, being an aprotic polar solvent, is more effective in extracting moderately polar compounds with antimicrobial activity. Additionally, acetone's lower boiling point compared to ethanol minimizes solvent residue in the extract, ensuring it does not interfere with the effectiveness of antimicrobial tests. Maceration was performed by weighing 50 grams of the ground sample and soaking it in 500 mL of solvent at a 1:10 ratio for 3×24 hours.

The carrots used in this study were large, fresh carrots. After being thoroughly washed, the carrots were thinly sliced and dried at room temperature for 7 days to prevent the degradation of bioactive compounds due

to high-temperature exposure. Once dried, the carrots were ground into a fine powder. Next, the maceration process was carried out using acetone as a solvent for antibacterial *(Escherichia coli)* and antifungal *(Candida albicans)* tests, and 96% ethanol as a solvent for the antioxidant test. The maceration was performed at a 1:10 ratio for 3 days. The use of different solvents for antimicrobial and antioxidant tests was based on the polarity properties of the compounds being extracted. Acetone, as an aprotic polar solvent, is more effective in extracting moderately polar compounds with antimicrobial activity. Additionally, acetone has a lower boiling point than ethanol, minimizing solvent residue in the extract and ensuring it does not interfere with the effectiveness of the antimicrobial test.

Phytochemical Screening

Flavonoids: A total of 5 mL of the extract sample was added with magnesium (Mg) ribbon and concentrated hydrochloric acid (HCl). A positive result is indicated by the formation of a yellow color. The reaction involved is as follows: Flavonoids + Mg + HCl → Flavilium compound (yellow color) (Ningsih et al., 2019).

Saponins: A total of 5 mL of the extract sample was diluted with distilled water and 96% ethanol, followed by the addition of 2N HCl and vigorous shaking. The formation of stable foam indicates a positive result. The reaction involved is: Saponins (triterpenoid/steroidal glycosides) + distilled water and 96% ethanol → stable foam (upon shaking).

Tannins: A total of 5 mL of the extract sample was treated with 5% ferric chloride ($FeCl_3$) solution. A positive reaction is indicated by the formation of a black-colored complex. The chemical reaction can be represented as: Tannins (phenolics) + Fe^{3+} → Iron–tannin complex (black) (Ningsih et al., 2019).

Alkaloids: A total of 5 mL of the extract sample was tested with Dragendorff's reagent. A positive result is indicated by the formation of an orange precipitate. The reaction is as follows: Alkaloids (R-N) + $[BiI_4]^-$ → [R-NH]BiI_4 (orange complex).

Antioxidant Test

Preparation of 0.3 mM DPPH solution. DPPH compound was weighed as much as 15.80 mg and dissolved in ethanol p.a 100 mL. Preparation of a vitamin C standard solution. Vitamin C powder was weighed as much as 10 mg and dissolved in ethanol p.a 100 mL. Then diluted into concentrations of 20, 40, 60, 120, 140 ppm. Each concentration of Vitamin C standard solution was pipetted as much as 1 mL and put in a test tube then added DPPH solution as much as 1 and 3 mL ethanol p.a. Homogenized and allowed to stand for 30 minutes in a dark room. Preparation of blank. A total of 1 mL of 0.3 mM DPPH was diluted with 4 mL of ethanol p.a then homogenized and incubated for 30 minutes. Antioxidant activity test. All solutions that have been made were measured for absorbance using a uv-vis spectrophotometer at a wavelength of 515 nm (Irfayanti et al., 2023).

Antibacterial and Anti-Fungal Test

The bacterial suspension of *Escherichia coli* and fungal suspension of *Candida albicans* were prepared by inoculating a single colony from a pure culture into sterile liquid media. *E. coli* was cultured in Nutrient Broth (NB), while *C. albicans* was cultured in Sabouraud Dextrose Broth (SDB). The cultures were incubated at 37°C for 18 hours for bacteria and 24 hours for fungi. After incubation, the microbial suspensions were standardized to 0.5 McFarland turbidity standard, equivalent to approximately 1.5×10^8 CFU/mL. These suspensions were then evenly spread on the surface of Mueller Hinton Agar (MHA) plates using the lawn culture technique.

For media preparation, 3.8 grams of MHA were dissolved in 100 mL of distilled water and sterilized. The extract was prepared at concentrations of 20, 40, 60, and 80%. The positive controls used were sulfamethoxazole for the antibacterial assay and ketoconazole for the antifungal assay.

The disc diffusion method was employed to evaluate antimicrobial activity. Sterile paper discs were soaked in each extract concentration and placed onto the surface of MHA plates previously inoculated with *E. coli* and *C. albicans*. The plates were then incubated at 37°C for 24 hours. Antimicrobial activity was determined by measuring the diameter of the inhibition zones using a vernier caliper, and the results were compared with those of positive and negative controls (Goetie et al., 2022; Widyasanti et al., 2016).

Result and Discussion

Based on the results of the research that has been done, the yield of carrots is 10.1% (v/w). Based on the yield that has been obtained, phytochemical screening is then carried out to determine the compounds contained in carrot extract. The results of phytochemical screening can be seen in Table 1.

Table 1. Phytochemical screening test results

Test Compound Result	Result
Flavonoids	+
Saponins	+
Tannins	+
Alkaloids	+

Based on Table 1, it is known that carrot extract contains flavonoids, saponins, tannins and alkaloid compounds. This is in accordance with the research of M & Rosa (2022) which states that carrots have secondary metabolite compounds in the form of flavonoids, saponins and alkaloids. Flavonoids are a group of polyphenolic compounds that function as antioxidants in plants. In carrots, flavonoids function to protect against UV damage and microbial infections. For humans, flavonoids in carrots are useful as antioxidants that fight free radicals, reduce the risk of heart disease, and have anti-inflammatory and anticancer properties. Saponins are compounds known for their ability to produce foam when reacting with water. In carrots, saponins function as a defense mechanism by fighting insects and pathogenic microorganisms. In humans, saponins have hypocholesterolemic (lowering cholesterol levels) and anticancer effects, and boost the immune system. Tannins are secondary metabolite compounds produced by carrots. Tannins are phenolic compounds that provide protection against plant-eating animals due to their bitter taste. In carrots, tannins function as natural antimicrobials that protect against pathogens. For humans, tannins have antioxidant and astringent properties, and can help reduce the risk of infection. Meanwhile, the alkaloid content in carrots acts as a chemical defense against insects and herbivores. Alkaloids found in carrots have therapeutic potential, such as anticancer, analgesic, and anti-inflammatory activities (Ahmad et al., 2019; Søltoft et al., 2010).

Characterisation using FT-IR was carried out at wavelengths of 3500-500 cm^{-1}. IR with intermediate wavelengths is an area used for the identification of organic compounds, because the absorption bands in this area are vibrations of certain functional groups. The results of characterisation can be seen in the Figure 1.

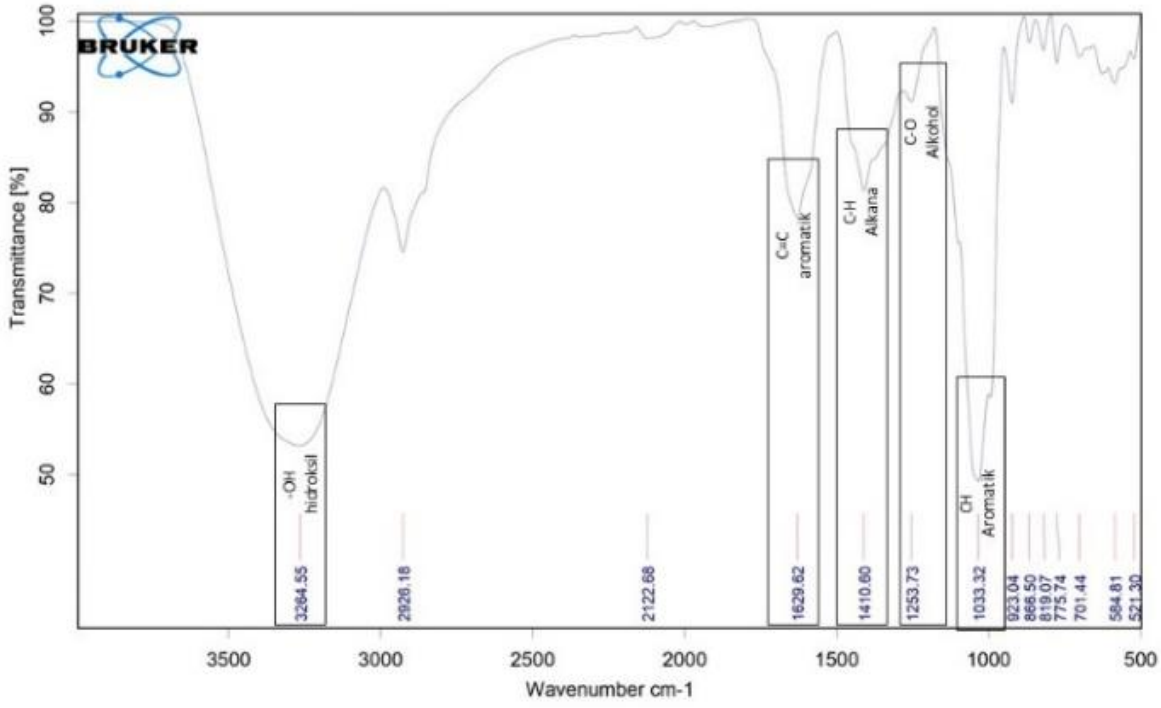

Figure 1. FTIR spectrum of carrots

Based on Figure 1, it shows the presence of -OH functional groups on the peak ramps formed at peak 3264 cm^{-1}. There is -C=C- aromatic at peak 1629 cm^{-1}, -C-H alkane group at peak 1410 cm^{-1}. These results indicate the presence of beta-carotene in carrots. The -C-O- alcohol group formed at peak 1253 cm^{-1}, and at peak 1033 cm^{-1} formed an aromatic -CH group. In general, these spectra show the presence of functional groups commonly found in carotenoids, flavonoids, and phenolic compounds, which correspond to carrot extracts, especially if carrots are extracted for compounds such as beta-carotene, which is one of the main bioactive components (Damayanti et al., 2021).

Antioxidant test results were carried out on extracts using 96% ethanol. This solvent was chosen because 96% ethanol has a level of solubility that can attract vitamin C and flavonoid compounds optimally. Antioxidant activity was measured using DPPH solution. The working principle of DPPH is the interaction between DPPH radicals and antioxidants contained in carrot extract. Antioxidants will transfer electrons or hydrogen H+ which will react with free radicals from DPPH. Based on the test results show that the greater the

concentration, the greater the inhibition power against DPPH free radicals. The antioxidant measurement results obtained based on the line equation formed is y = 0.2214x + 22.116. R2 = 0.9878, then the IC_{50} value of 125.944 ppm was obtained. Based on the research of Zongo et al. (2023) showed that carrot extract has a weak antioxidant content (101-150 ppm).

The next test is an antibacterial test using gram negative bacteria, namely *Escherichia coli (e. coli)*. The control used in this study was sulfamethoxazole. The test results are shown in Table 2.

Table 2. Test results of inhibition zone diameter measurement on *E. coli* Bacteria

Replay	Diameter of inhibition zone (mm)			Control
	40%	60%	80%	
E. coli 1	0.70	0.80	0.90	2.73
E. coli 2	0.70	0.74	0.80	2.73
Average	0.70	0.77	0.85	2.73

Based on the test results, it is known that the greater the concentration of carrot extract used, the greater the inhibition. All three concentrations showed moderate inhibition (5-10 mm). The inhibitory power given by carrot extract is due to the presence of active compounds in carrots, namely flavonoids, saponins, tannins and alkaloids that act as antibacterials. Flavonoids are known to disrupt bacterial cell membranes and inhibit bacterial enzymes. This can occur because flavonoids can denature the enzymes in these bacteria. Flavonoids also have the ability to form complexes with soluble extracellular proteins and with cell walls, so that microorganisms cannot attach and invade cells. Flavonoids are also able to release transduction energy to the bacterial cytoplasmic membrane and inhibit bacterial motility (Manik et al., 2014). In addition, flavonoids can also cause bacterial cell wall damage through inhibition which results in the incorporation of non-cross-linked glycan chains into the peptidoglycan of the cell membrane so that it becomes one weak structure (Donadio et al., 2021). Meanwhile, saponins can cause leakage of intracellular components and inhibit bacterial growth. The mechanism of saponins as antibacterial is by damaging the bacterial cell membrane due to an increase in membrane permeability due to saponins interacting with the bacterial cell wall (Eylands et al., 2021; Kakarla, 2009). Tannin compounds can inhibit bacterial growth by binding bacterial proteins or enzymes. While alkaloid compounds are known to damage the structure of bacterial cell membranes (Javed et al., 2020; Yan et al., 2021). The next test conducted was the antifungal test. Based on the antifungal test, the results are shown in Table 3.

Table 3. The test results of measuring the diameter of the inhibition zone on *Candida albicans* fungus

Replay	Diameter of Inhibition zone (mm)			Control
	60%	80%	100%	
Candida albicans 1	-	-	0.830	2.700
Candida albicans 2	-	-	0.840	2.700
Average	-	-	0.835	2.700

The results of the antifungal test were carried out by calculating the diameter formed using a vernier scale. The inhibition zone formed was at 100% extract concentration, where the concentrated extract was used as an antifungal without being diluted first. Antifungal compounds have various inhibitory mechanisms against fungal cells. One of them is neutralising enzymes associated with fungal invasion, damaging fungal cell membranes, inhibiting fungal enzyme systems thus disrupting hyphae formation and affecting the synthesis of nucleic acids and proteins. Flavomoid secondary metabolite compounds can cause inhibition of the enzyme glucan synthase which plays a role in the formation of fungal cell walls. In addition, fungal inhibition patterns can also occur because tannin compounds can denature proteins, which causes damage to the integrity of fungal cells. The next secondary metabolite compound that can damage the fungal cell membrane is saponin. Saponins function as antifungals by increasing membrane permeability. This will cause leakage of the fungal cellular components (Datta et al., 2022; Sharma & Sharma, 2022).

Conclusion

The ethanol extract of carrot (*Daucus carota* L.) yielded 10.1% and demonstrated moderate antioxidant activity with an IC_{50} value of 125.9 ppm, indicating its potential to neutralize free radicals. Phytochemical screening confirmed the presence of flavonoids, tannins, saponins, and alkaloids, which are likely responsible for the observed activity. The extract also exhibited antibacterial activity against *Escherichia coli*, with the highest inhibition zone of 0.85 mm at 80% concentration, and antifungal activity against *Candida albicans*, with the highest inhibition zone of 0.835 mm at 100% concentration. These findings support the research objective to assess the antioxidant and antimicrobial potential of carrot extract and suggest its possible application as a natural health-promoting agent.

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Conceptualization, formal analysis, data curation, D.R.P.; methodology, validation, D.P.; investigation, resources, L.M.; writing—original draft preparation, S.MF.S. and K.B.S.; writing—review and editing, W.H. and S.F.M.; visualization, W.H.; Translator, S.F.M. All authors have read and agreed to the published version of the manuscript.

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The authors declare no conflict of interest.

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