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Antioxidant Activity of Single Black Garlic (*Allium sativum L.*) Ethanol Extract and Its Potential for Oral and Dental Health

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Abstract:

Allium sativum L., or fermented single garlic, is an garlic that is heated for 14 days at 70°C with a humidity of 70–80% to turn it black. In this study, the total phenolic content and antioxidant activity of an ethanol extract from a single black garlic (*Allium sativum* L) are to be determined. Maceration is used to extract a single black garlic (*Allium sativum* L), and the folin-ciocalteau method is used to measure total phenolics. Using the 1,1-diphenyl-2-picrylhydrazyl (DPPH) technique, antioxidants were evaluated. The results of the study demonstrate that the ethanol extract of a single black garlic (*Allium sativum* L) has a total phenolic content of 62,45% ± 0,2886 and that the extract's antioxidant activity has an IC50 value of 10.4156 µg/mL (very strong antioxidant). The high phenolic content and antioxidant activity which is classified as very strong indicate that single black garlic ethanol extract has potential for oral and dental health.

Keywords: *Allium sativum* L, Single Black Garlic, Total Phenolic, Antioxidant, Ethanol Extract

1. Introduction

The field of disease prevention has expanded in recent years, with a particular focus on the function of antioxidants and free radicals. The potential roles of "free radicals" in diseases and "antioxidants" in their prevention are compiled in this review. Particular attention is given to the present state of antioxidants in oral disorders as well as their potential future applications in dentistry. Compounds called antioxidants are utilized by aerobic organisms to fend against oxidative stress, which is brought on by free radicals and active oxygen species. By preventing the production of free radicals or eliminating them, they provide protective functions. According to Satyanarayana and Chakrapani (1999) and Papas (1999). Research has shown that a number of biological effects can prevent the development of cancer. These encompass impacts on tumor start, development, and advancement, cell proliferation and differentiation, immunological function, DNA repair, and cell membrane stability (Radler, 2003; Schiferle, 2005). A lot of attention has been paid to dietary antioxidants as possible cancer chemopreventive agents, including carotenoids, vitamins C and E, and selenium.

Garlic, or Allivum sativum L. in scientific parlance, is a natural component that can be utilized. Due to its allicin content, single garlic has been shown in several studies to have a number of health benefits, including antibacterial properties (Pudiarifanti & Farizal, 2022). However, research on single garlic is still scarce in the literature. A single garlic bulb (*Allium sativum* L) has comparatively higher nutritional quality than an onion

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bulb with several bulbs. Antioxidant chemicals found in single garlic (*Allium sativum* L) have the ability to neutralize free radicals (Wibisono et al., 2020).

Alicin, an organosulfur molecule found in single garlic cloves, is what gives them their pungent scent and gives them pharmacological effect (Dewi et al., 2021). It is known that the phenolic chemicals and flavonoids found in single garlic bulbs have antioxidant properties (Januarti et al., 2019). According to Deanita et al. (2022), solitary garlic is recognized to have more antioxidant content than ordinary garlic. Strong antioxidants found in a single clove of garlic, known as flavonoids, can lower blood triglyceride levels and lower the risk of degenerative diseases when taken frequently.

Figure 1. Single Garlic and Single Black Garlic (*Allium sativum L.*) (Abdurrahman et al., 2022; Pramita & Yani, 2020).

Garlic has several recognized pharmacological benefits, including antibacterial, antihypertensive, anticholesterol, and antioxidant properties (Azhar & Yuliawati, 2021). Antioxidants, also known as free radical trapping compounds, are chemicals that have the ability to neutralize free radicals or that work to shield the biological system of the body from harmful effects resulting from reactions or processes that produce excessive oxidation. According to Fessenden & Fessenden (1986), free radical compounds are produced by the body through a variety of intricate chemical processes. These processes include breathing, cell metabolism, excessive exercise, inflammation, and exposure to environmental pollutants like chemicals, smoke from cigarettes, motor vehicle fumes, and sunlight.

Black garlic is the result of processing garlic through a heating process with a temperature of 70°C and 70-80% humidity for 30-40 days without the addition of other substances or any additional treatment. Black garlic has a strong antioxidant content (Kimura et al, 2017). Antioxidants are compounds that can inhibit oxidation reactions by binding to free radicals and highly reactive molecules (Winarsi, 2007). Several studies have shown that black garlic has antioxidant activity which is very useful for preventing several diseases, including inhibiting the growth of cancer cells and anti-allergy preventing premature aging anti-obesity, lowers blood lipid levels, and lowers blood sugar levels (Kimura et al., 2017),

Based on research conducted by Choi et al., (2014) black garlic extracted with deionized water was prepared for 7, 14, 21, 28 and 35 days under controlled conditions of 70°C and 90% relative humidity. The results of this study showed an increase in antioxidant activity in black garlic occurred at a maximum when heated for 21 days, the results of these tests on fresh garlic and black garlic had antioxidant activity of 4.65% and 74.48%. the antioxidant activity test of black garlic ethanol extract with a heating time of 12 days, the black garlic ethanol extract obtained an IC50 value of 637.7955 µg/mL (very weak antioxidant). In research Azizah (2024) findings that the ethanol extract of black garlic had an overall phenolic content of 8,96 % and IC50 value of 4.7527 µg/mL (extremely strong antioxidant) for its antioxidant activity with soxhletation method for extract black garlic. Based on the description above, researchers are interested in examining the antioxidant activity of single black garlic for 14 days and its potential for dental and oral health.

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2. Data Collection Methods

Tools and Materials

The equipment used includes: Double Beam UV-Vis Spectrophotometer (Shimadzu UV-1800), rotary evaporator (BUCHI Rotarspor R 200®), cuvette, pipette volume, analytical balance (Prescisa), rice cooker (miyako), blender (miyako), ThermoHygrometer (Brand), vials, furnace (Carbolite Gero), and glassware laboratory.

The ingredient used in this research was single garlic (Allium sativum L) as much as 1 Kg. The chemical used is 70% Ethanol (C_2 H_5 OH) (PT. Novalindo), Distilled Water (H_2 O) (PT. Novalindo), Acetic Acid Anhydrous P (CH_3 $CO)_2$ O) (Merck), Mg powder (brand), Sulfuric Acid (H_2 SO_4) (Merck), Potassium Iodide (KI) (Merck), Iodine (I_2) (Merck), and Iron (III) Ammonium Sulfate (Merck), Hydrochloric Acid (HCl) (Merck), Mercury (II) Chloride (Hg_2 Cl_2) (Merck), Chloroform ($CHCl_3$) (merck), distilled water (PT. Novalindo), Methanol (CH_3 OH) p.a (merck), Mayer's Reagent (HgI_4K_2) (Merck), vanillin (brand), gellatin (Merck), Bouchardat's Reagent (Merck), 1,1-Diphenyl-2-Picrilhydrazil (DPPH) (Sigma) Gallic Acid (C_7 H_6 O_3) (PT. Bratachem), Folin-Ciocalteu (Merck)

3. Analytical Method

Procedures

One kilogram of raw garlic (*Allium sativum L.*) is used as the sample. Located in the West Sumatra Province's Pasar Raya Padang, Padang City. In the Department of Biology, Faculty of Mathematics and Natural Sciences (FMIPA), Andalas University (UNAND) Padang, West Sumatra, single garlic was identified in the Andalas University Herbarium.

Procedure to Make a Single Black Garlic

1 kg of whole garlic, wrapped in aluminum foil, placed inside the rice cooker, sealed, placed on maintain warm, and left for 14 days. 14 days after it was heated. Once fermentation is complete, remove the single garlic (Allium sativum L), weigh the onion single white (Allium sativum L), peel the onion skin, and blend (Azizah et al, 2020)

Making A Ethanol Extract From Single Black Garlic.

200 g of single black garlic are combined with 2000 mL of 70% ethanol solvent (ratio 1:10 w/v) in a maceration bottle. Soak it for 6 hours, stirring from time to time, and then leave it for 18 hours. The maceration procedure is done again with the same type and quantity of solvent, separating the macerate by filtering. A thick extract is produced by collecting all of the macerates and then evaporating them in a rotary evaporator at temperatures lower than 40–50ºC. Finally, determine the yield (Azizah et al., 2020).

Single Black Garlic Ethanol Extract: A Qualitative Analysis (*Allium sativum* L).

1. Test for Flavonoids
   A total of 0.5 g of single black garlic ethanol extract (*Allium sativum* L) was dissolved with 3 mL of 96% ethanol, then the powder was added Mg 0.1 g and 2 drops of concentrated HCl. Positive results are indicated by the appearance of color orange yellow to dark red (Ministry of Health of the Republic of Indonesia, 1995).
2. Alkaloid Examination
   Add 1 mL of 2 N hydrochloric acid and 9 mL of distilled water to 0.5 g of single black garlic ethanol extract (*Allium sativum* L), heat over a water bath for two minutes, cool, and strain. Testing for alkaloids is done using the resulting filtrate. Add the filtrate (0.5 ml). To create a brown to black precipitate, add two drops of Wagner LP to each test tube. A white or yellow precipitate is formed by two drops of the Mayer LP reagent. A crimson precipitate is produced by two drops of the Draeger-Larkin LP reaction (Department of Health of the Republic of Indonesia, 1995).
3. Test for Saponin
   Fill a test tube with 0.5 g of *Allium sativum* L. single black garlic ethanol extract, followed by 10 ml of distilled water. After that, shake briskly for ten seconds. If foam appears, it will persist for at least 10 minutes, measure 1 to 10 cm in height, and not vanish even after adding one drop of HCl 2 N (Ministry of Health of the Republic of Indonesia, 1995).

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4. Tannin Examination
   Allium sativum L. single black garlic ethanol extract (0.5 g), added with 10 mL of hot distilled water, 3 drops of 10% NaCl, stirred, and filtered. Gelatin was then added, along with 5 ml of 10% NaCl.
5. Terpenoids and steroids Test
   A total of 0.5 g of single black garlic (*Allium sativum* L) ethanol extract. adding 2 milliliters of concentrated sulfuric acid (H_2SO_4) and 2 milliliters of anhydrous acetic acid, along with 3 milliliters of chloroform or 3 milliliters of 96% ethanol. The presence of steroid molecules and their production are indicated by a color shift from purple to blue or green. Terpenoid chemicals are present because of the brownish tint that exists between the surfaces. (Ministry of Health Republic of Indonesia,1979).
6. Phenol Test
   A total of 0.5 grams of single black garlic ethanol extract (*Allium sativum* L), add 10% vanillin P solution in 90% ethanol, then add 2 drops of hydrochloric acid P, the part containing phenol is colored red intensive (Ministry of Health of the Republic of Indonesia 1995).

Quantitative Analysis of Single Black Garlic Ethanol Extract (*Allium sativum* L)

1. Phenol

a. Making gallic acid stock solution (100 µg/mL).
   Carefully weighed 10 mg gallic acid, then dissolved in methanol p.a ad to a volume of 100 mL (100 µg/mL).
b. Maximum wavelength (60 µg/mL).
   A gallic acid solution was prepared with a concentration of 60 µg/mL by means of Pipette 6 mL of µg/mL gallic acid solution into a volumetric flask 10 mL. Then pipette 1 mL of a solution of 60 µg/mL concentration into the vial add 5 mL of Folin-Ciocalteu dilution (7.5% in water). Leave for 8 minutes, add 4 mL of 1% NaOH, incubate for 1 hour at room temperature. Determine the maximum wavelength of gallic acid using UV-Vis spectrophotometry at a wavelength of 400-800 nm (Ministry of Republic of Indonesia Health, 2011)
c. Making gallic acid calibration curve
   Solutions were made with concentrations of 20, 40, 60, 80, 100 µg/mL by Pipette 2, 4, 6, 8, 10 mL of 100 µg/mL gallic acid solution, put into 10 mL volumetric flask, added with methanol p.a to the mark. From 1 mL of gallic acid solution was pipetted into the vial for each concentration. Add 5 mL of Folin-Ciocalteu solution (7.5% in water). Leave for 8 minutes, add 4 mL of 1% NaoH, incubate for 1 hour at room temperature. Determine the absorbance at the maximum wavelength with a spectrophotometer UV-Vis. Next, determine the regression equation and calibration curve (Ministry of Health of the Republic of Indonesia, 2017)
d. Calculating the total phenolic content of a single black garlic ethanol extract (*Allium sativum* L).
   In a 100 mL volumetric flask, weigh out 10 mg of single garlic ethanol extract (*Allium sativum* L), ferment it, and then gradually add methanol until the flask reaches the limit. Subsequently, 6 mL of the stock solution (100 µg/mL) was pipetted to create a solution with a concentration of 60 µg/mL. Fill a 10-milliliter measuring flask halfway full with methanol. Transfer 5 mL of the 7.5% in water Folin-Ciocalteu solution using a pipette to 1 mL in the vial. Add 4 mL of 1% NaOH and incubate for 8 minutes at room temperature after letting stand for 8 minutes.

Making a stock solution of 30 µg/mL DPPH

Approximately 10 mg of DPPH were carefully weighed (BM 394.33). after which it was dissolved to a volume of 100 mL using methanol p.a. and put into an aluminum foil-covered measuring flask. Sufficient solvent to meet the mark, then shake until completely blended and a 100 µg/mL DPPH solution is formed. then 15 mL of strong DPPH solution was pipetted to dilute it. To make a DPPH solution with a concentration of 30µg/mL, put 100 µg/mL in a 50 mL volumetric flask, add enough solvent to reach the mark, and shake until homogenous (Molyneux, 2004).

Creating Blank Solutions for Optimal Wavelength

Pipette DPPH Fill the container with 3.8 mL of DPPH 30 solution (µg/mL). After adding up to 0.2 mL of methanol p.a. and homogenizing the mixture, the vial was sealed with aluminum foil. then spent 30 minutes incubating in a dark room. A UV spectrophotometer can be used to determine the absorption wavelength. observable between 400 and 800 nm in wavelength, and ascertain the maximum wavelength (Molyneux, 2004).

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Gallic Acid Stock Solution Preparation

After weighing 10 mg of pure gallic acid and adding 100 µg/mL of methanol p.a. to the flask, homogenize the mixture (Andayani et al., 2008).

Gallic Acid Calibration Curve Preparation.

A concentration series of 4 µg/mL, 6 µg/mL, 8 µg/mL, and 10 µg/mL was then created. 0.4 mL of the stock solution (100 µg/mL), 0.6 mL, 0.8 mL, 1 mL, and 1.2 mL were pipetted, placed in a 10 mL measuring flask, and then filled with methanol p.a. to the limit mark. Then, pipette 0.2 milliliters. Fill a vial with 3.8 mL of DPPH (30 µg/mL) solution for each concentration, and cover the vial with aluminum foil. 30 minutes were spent incubating in a dark room. A UV-Visible spectrophotometer operating at maximum DPPH wavelengths was used to evaluate the absorbance of different concentrations (Andayani et al., 2008).

Measurement of Antioxidant Activity of Single Black Garlic Ethanol Extract (*Allium sativum* L)

Weighed the ethanol extract of single black garlic (*Allium sativum* L) 10 mg, then dissolved with methanol p.a in a flask Measure 100 mL, add enough to the limit mark to get a concentration of (100 µg/mL). then do the dilution by adding methanol p.a so that samples with concentrations of 4, 6, 8, 10, 12 µg/mL are obtained by means pipette 0.4 mL; 0.6 mL; 0.8 mL; 1 mL; 1.2 mL of 100 µg/mL, respectively put into a 10 mL volumetric flask and add methanol p.a to the mark limit. Antioxidant activity in sample solutions and solutions is measured. To provide a comparison, 0.2 milliliters of each concentration were pipetted, then 3.8 milliliters of 30 µg DPPH solution were added using a micropipette. The mixture was then placed into a vial and covered with aluminum foil. After homogenizing the mixture, it was allowed to sit in a dark area for half an hour. Utilizing UV-Visible spectrophotometry, calculate absorbance on 516 nm is the maximal wavelength of DPPH (Andayani et al., 2008).

IC50 determination

The percentage of inhibition of DPPH radicals from the sample solution can be calculated using the formula:

$$\%\ Inhibition = \frac{Control\ Absorbance - Sample\ Absorbance}{Control\ Absorbance}\ x\ 100\%$$

Information:

% Inhibition	: Percentage of antioxidant activity power
Control absorbance (A1)	: DPPH absorbance 30 µg/mL
Sample absorbance (A2)	: Sample absorbance test

After obtaining the percentage of inhibition from each concentration, then the percent of inhibition is plotted on the x and y axes respectively in the linear regression equation y = a + bx. This equation can be used to determine the IC50 value with the formula (Andayani *et al*., 2008).

$$IC50 = \frac{50 - a}{b}$$

RESULTS AND DISCUSSION

One kilogram of single garlic bulbs (*Allium sativum* L), bought at Pasar Raya Padang, Kota Padang, West Sumatra Province, served as the study's sample. Plant identification has been done at the Andalas University (UNAND) Padang, Sumatera West Herbarium, Department of Biology, Faculty of Mathematics and Science, Natural Sciences (FMIPA). Finding out the identity of the sample that will be used is the goal of sample identification. These findings demonstrate that a single garlic bulb (*Allium sativum* L), a member of the Amaryllidaceae family, was the sample used in this study. Whole bulbs of single garlic (*Allium sativum* L) are processed into single garlic (*Allium sativum* L) fermented by means put in a rice cooker with a temperature of 70º C and humidity 76-80% measured by inserting an indicator or sensor into the Thermohygrometer into a rice cooker lined with aluminum foil. then fermented for 14 days with the rice cooker set to inside keep warm mode. Ferment single garlic (*Allium sativum* L) for 14 The day was chosen because it is the optimal time to obtain the chemical content on single garlic (*Allium sativum* L). If the fermentation time is too long for a long time, it will reduce the quality of black garlic due to the enzyme contained in black garlic is very sensitive and unstable to changes in temperature, This can trigger damage to black garlic (Azhar & Yuliawati, 2021) *Allium sativum L.*, or single garlic, during its fermentation process.

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Single garlic (*Allium sativum* L) changes to single black garlic (*Allium sativum* L) in terms of weight, color, texture, taste, and aroma. 884.8 grams of single garlic extract (*Allium sativum* L) were obtained following fermentation. Because volatile chemicals are affected by temperature and time of fermentation, a drop in weight of 115.8 grams suggests a decrease in water content. The color change that occurs is from white to yellow, then light brown, dark brown until finally black. Texture of Single garlic (*Allium sativum* L) turns soft and watery like into single black garlic (*Allium sativum* L). These changes occurs as a result of fiber bonds breaking down into simple sugars.

The maceration process is then used to extract a single black garlic (*Allium sativum* L). To do this, 200 grams of garlic were weighed, and the garlic was extracted using a 70% ethanol solvent for three times in a 24-hour period. The maserate is a liquid extract, which is then used to separate the extract from the filter liquid using a rotary evaporator. The heating is aided by the pressure decrease and the round-bottom flask's rotation, which might cause evaporation. The vapor from the filter solution rises to the condenser with the aid of a vacuum pump, where it condenses into liquid molecules of pure solvent and is collected in a round-bottom flask to yield a thick extract from fermented single garlic. Yield from fermented ethanol extract of single black garlic (*Allium sativum* L) is as much as 66.83%.

The next test is a phytochemical screening test to find out content of secondary metabolite compounds contained in ethanol extract single black garlic (*Allium sativum* L). On ethanol extract single black garlic (*Allium sativum* L) showed positive results on alkaloids, flavonoids, phenols, tannins and terpenoids. Judging from the positive alkaloid test results the chemical reaction between Mayer's reagents forms a white or yellowish precipitate, Wagner's reagent forms a brown precipitate and Dragendroff's reagent forms red precipitate. A positive flavonoid test occurred when a chemical reaction occurred between the Mg powder reagent and concentrated HCl forms an orange/dark red color. A positive phenol test occurs the chemical reaction between 10% vanillin P solution and HCl forms a red color intensive. The results of the tannin test were positive, a chemical reaction occurred between the 10% NaCl reagent and the gelatin solution forms a white precipitate. The saponin test was negative for foam formed less than 1 cm after adding hot water and then shaking it plus 1 drop of HCl. In the terpenoid test, a chemical reaction occurs between the chloroform reagent, H_2 SO_4 and anhydrous acetic acid with a brownish ring indicate positive Terpenoids.

Gallic acid was used as a standard solution in the Folin-Ciocalteu method to determine the total phenolic content of the single black garlic ethanol extract (*Allium sativum* L). Gallic acid is categorized as an acid simple phenol, a derivative of hydroxybenzoic acid (Kupina, et al., 2018). By using a Double Beam UV-Vis Spectrophotometer to detect the content of gallic acid at 60 µg/mL, the highest absorption wavelength was found to be 765.50 nm, with an absorbance of 0.568.

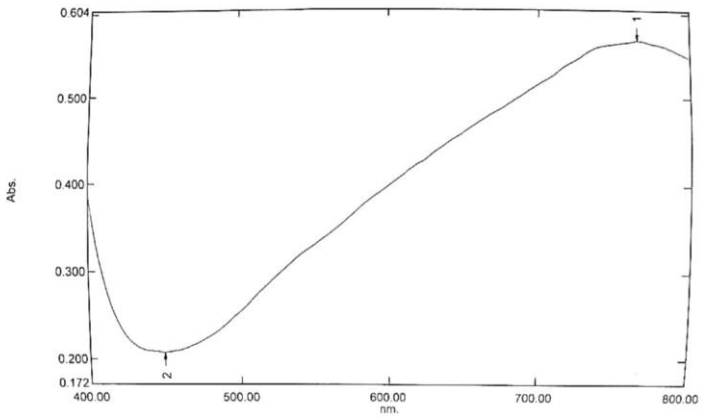

Figure 3. Maximum Absorption Wavelength of Gallic Acid Concentration 60 µg/mL.

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The results of determining gallic acid phenolics as standard phenolic content at a maximum absorption of 765.50 nm with a concentration of 20, 40, 60, 80, and 100 µg/mL obtained a solution absorbance of 0.397; 0.465; 0.530; 0.601; 0.668, obtained by the equation y = 0.00339 x + 0.3288 with the regression coefficient 0.99993. The greater the concentration of the compound phenolic, the more phenolic ions are formed. So, it will increase much of it is reduced by phenolic ions and the resulting blue color increases concentrated (Lee et al., 2003). Results of measuring the total phenolic content of single black garlic ethanol extract (*Allium sativum* L) fermentation with a concentration of 60 µg/mL was measured using UV-Vis spectrophotometer at the maximum wavelength of gallic acid 765.50 nm, the absorbance obtained with 3 repetitions was 0.534, 0.533, 0.534 respectively. each absorbance is subtracted from the blank absorbance, namely 0.078, to obtain a percent average total phenolic content 62.45% ± 0.2886. Phenolic compounds have oxidation-reduction properties that work as antioxidants because, when they react with free radical compounds, the hydroxyl group on a functional aromatic ring acts as a hydrogen atom donor, slowing down the oxidation process and reducing the free radical compounds. Accordingly, the antioxidant activity of phenolic compounds increases with the number of hydroxyl groups present (Lee et al., 2003).

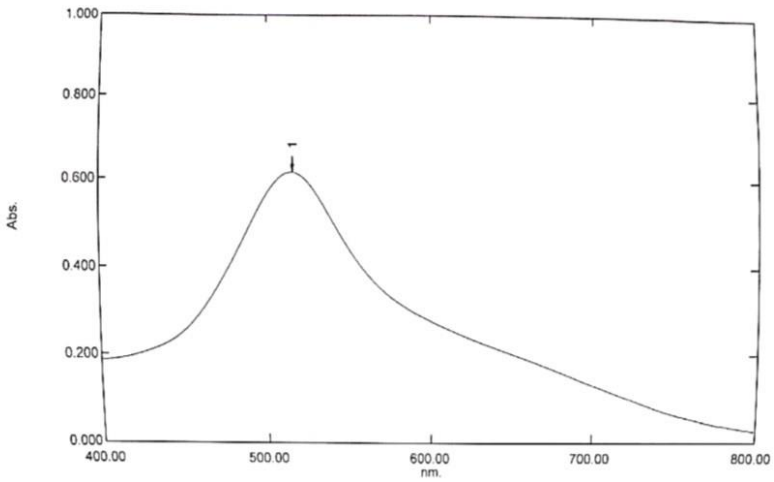

Figure 4. Absorption Wavelength Maximum DPPH Concentration 30 µg/ mL

Using gallic acid as a reference, the antioxidant activity test produced solution absorbance values of 0.476, 0.414, 0.348, 0.281, and 0.219 as well as percent activity radical antidote values of 22.8525%, 32.9011%, 43.5980%, 54.4570%, and 64.5056%. Based on the absorbance results, it is evident that lower absorbance values are obtained at higher sample concentrations. This is because higher concentrations of antioxidant compounds are more effective at reducing or warding off radicals in DPPH, leading to larger inhibition percentages (Bahriul et al, 2014). The gallic acid comparison yielded an IC_{50} value of 9.2086 µg/mL, or 50% free radical scavenging activity (extremely potent antioxidant <50 µg/mL).

Table 1. Results of measurement of the absorbance of DPPH + ethanol extract of Black Garlic at a wavelength of 516.00 nm and an absorbance of 0.617 with a double beam spectrophotometer.

No	Concentration(µg/mL)	Absorbance		% inhibition	IC_{50}
		A1	A2		
1.	4	0.617	0. 503	18.4764%	10.4156 µg/mL
2.	6		0.443	28.2009%	
3.	8		0.380	38.4116%	
4.	10		0.320	48.1361%	
5.	12		0. 262	57.5364%	

When single black garlic (*Allium sativum* L.*)* ethanol extract was tested for antioxidant activity, the absorbance was found to be 0.503, 0.443, 0.380, 0.320, and 0.262. The percentage of radical scavenging activity was found

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to be 18.4764%, 28.2009%, 38.4116%, 48.1361%, and 57.5364%. Based on the absorbance results, it can be observed that a higher sample concentration corresponds with a smaller absorbance value. This is likely due to the higher concentration of antioxidant compounds in the sample, which can reduce or ward off radicals in DPPH and increase the percentage of inhibition (Bahriul et al, 2014). From single black garlic ethanol extract an IC_{50} value or free radical scavenger activity of 50% was achieved (10.4156 µg/mL) (extremely antioxidant strong <50 µg/mL).

Research has shown that a single black garlic ethanol extract has a very high level of antioxidant activity, suggesting significant benefits for oral and dental health. Research Malcangi et al (2023) From the qualitative analysis of the articles included in the review, the oral diseases investigated for the evaluation of the potential beneficial effect of natural antioxidants are periodontal disease, mucositis, oral submucosal fibrosis, candidiasis, caries, oral lichen planus (OLP), and oral potentially malignant disorders (OPMDs) (Figure 5).

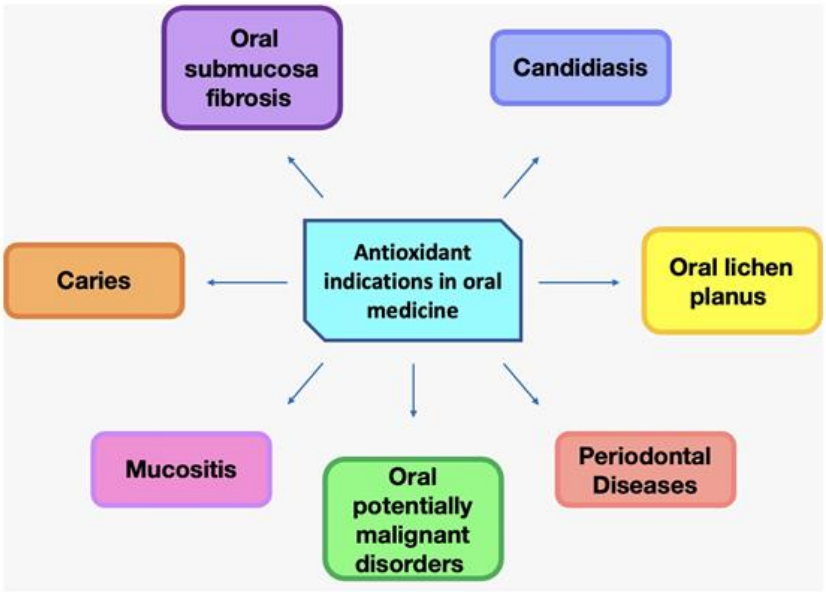

Figure 5. Oral Diseases Affected by Antioxidant Intake (Malcangi et al, 2023)

Periodontal Health

Periodontitis is a microbiological challenge to the immunoinflammatory host response caused by the dental biofilm, which leads to the physiologically accelerated degradation of connective tissue and bone. Since arachidonic acid metabolites, pro-inflammatory mediators, and ROS are involved in pathogenesis, pharmaceutical inhibition is advised as a substitute tactic Chapple (2009). Research has shown that vitamin C, a reducing agent that is soluble in water and provides electrons, helps to keep cells' redox potential in balance by scavenging reactive oxygen species (ROS) produced by oxidative stress and the subsequent inflammatory reactions Dawson et al. (2014) (Figure 6).

Figure 6. Redox Mechanism In Pathological And Physiological Cellular Metabolism.

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A grape seed proanthocyanidin extract and other commercial polyphenols (such as monomeric polyphenols, gallic acid, and epigallocatechin 3-gallate) were tested for their potential effects on the production of reactive oxygen species (ROS) and reactive nitrogen species (RNS) as well as on the expression of the inducible nitric oxide synthase protein in murine macrophages stimulated with lipopolysaccharides (LPS) from periodontopathogens. According to the study, proanthocyanidins are an effective therapeutic agent for the prevention of periodontal diseases and have strong antioxidant characteristics. Rich and naturally occurring in vitamins and minerals, black raspberries include a phytochemical. Black raspberries are particularly rich in various anthocyanins and phytosterols, as well as vitamins A, C, and E, folic acid, calcium, selenium, ß-carotene, ellagic acid, coumaric acid, and quercitin. In mice, freeze-dried black raspberries inhibit the growth of oral, esophageal, and colon cancer; in humans, trials have demonstrated a reduction in the expression of molecular dysplasia indicators. Human gingival fibroblasts activated with lipopolysaccharide (LPS) produce less nitric oxide thanks to the action of several flavonoids, including luteolin, quercetin, and genistein.

The Southeast Asian plant Camellia sinensis is the source of Green Tea (GT) extract, which is used to treat periodontal disease and dental cavities. Sur et al (2017). It is rich in PFs known as catechins, which include epigallocatechin-3-gallate (EGCG) and epigallocatechin (EGC) antioxidant substances capable of counteracting inflammatory processes and reducing the production of the main pro-inflammatory cytokines by the inflamed periodontal tissue (IL-1, IL-6, PGE2, and tumour necrosis factor alpha (TNF-alpha)), which are at the root of the onset of gingival infections and caries processes (Narotzki et al. (2012); Inchingolo et al. (2018); Malcangi et al. (2022). Because catechins, particularly EGCG, have antimicrobial properties, they prevent the disease from progressing. As a disinfectant, GT infusion works as a mouthwash and treats halitosis by lowering the quantity of volatile sulfur compounds that bacteria make when they interact with food residues, salivary proteins, and epithelial cells. It enhances the healing process when applied following scaling and root smoothing procedures. Its abundance of antioxidants prevents cell damage by causing oral cancer cells to undergo apoptosis, lowering the risk of mouth cancer and delaying its development even among smokers. et al. Koh (2011). It has been established that GT has a chemopreventive and therapeutic role in tumors of the oral cavity, particularly squamous cell tumors. It has been observed to alter the expression of genes in oral tumors, decrease the activation of phase I enzymes such as DT-diaphorase (DTD), arryl hydrocarbon hydroxylase (AHH), cytochrome b5, cytochrome P450, cytochrome b 5 reductase, and increase phase II enzymes such as glutathione-S-transferase (GST) and UDP-glucuronyl transferase (UDP-GT) (Zigmundo, et al., 2022); Srinivasan, et al (2008).The continual injection of GT PFs has demonstrated the challenge of distinguishing precancerous phases from cancerous lesions. Changes in cell membranes, epigenetic variables affecting DNA, RNA, and protein transcription systems, and the capacity of tea polyphenols to affect cell activity could all be involved in this Sur et al. (2017). In dental implant surgery, GT promotes bone integration processes by lowering blood cholesterol levels, the absorption of unsaturated fats, hyperlipidemia, the onset of osteoporosis, and the amount of bacteria in the oral cavity (Dundar et al.(2020); Keuroghilan et al., (2015))

The results of the study demonstrate that the ethanol extract of a single clove of garlic (*Allium sativum* L) fermentation has a total phenolic content of 62,45% ± 0,2886 and that the extract's antioxidant activity has an IC50 value of 10.4156 µg/mL (very strong antioxidant). The antioxidant activity with the DPPH method obtained IC50 the ethyl acetate fraction, n-hexane fraction and ethanol extract respectively of 31.6818 µg/mL, 194.5385 µg/mL and 6.6000 µg/mL (Azizah, et al 2022). According to research findings, because single black garlic has high phenolic levels and antioxidant activity, single black garlic ethanol extract may benefit dental and oral health.

CONCLUSIONS

The results of the study demonstrate that the ethanol extract of a single clove of garlic (*Allium sativum* L) fermentation has a total phenolic content of 62.45% ± 0.2886 and that the extract's antioxidant activity has an IC50 value of 10.4156 µg/mL (very strong antioxidant). Research findings indicate that because single black garlic contains high phenolic levels and antioxidant activity, single black garlic ethanol extract may benefit dental and oral health.

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There is growing evidence from studies on antioxidant combinations that these products could be beneficial in treating dental diseases and preserving oral health. Over the past few years, it has been increasingly clear how important it is to comprehend the advantages that nature offers in order to develop new strategies and resources to address the problems that medicine faces. Studies are being carried out on natural substances that have the potential to enhance human health via various biochemical processes that support the maintenance of the balance between ROS and antioxidants. The field of study that examines all of the components or active substances in food that have a positive impact on health, sickness prevention, and treatment is known as "nutriceutics," a combination of the words "nutrition" and "pharmaceutics." Organic or synthetic antioxidants have the potential to prevent many diseases at an early stage and function best when present in high concentrations. However, it is important to acknowledge that, at this time, larger case studies and cross-sectional studies are needed to further investigate and elucidate the seemingly contradictory findings of some of the studies we have reviewed. It is also crucial to stress how basic research and clinical/dental feasibility are inextricably linked and how new discoveries may be made by studying the "microenviroment" in which these molecular pathways operate.

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