Palestinian Medical and Pharmaceutical Journal

Exploring Antioxidant, Antibacterial, and Toxicity Prediction of Purslane Herb (Portulaca oleracea L.) from Several Extraction Solvents

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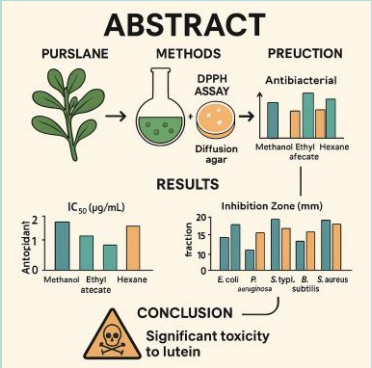
Received: 30th May 2024, Accepted 25th Apr. 2025, Published××××, , DOI: https://doi.org/10.xxxx

Abstract: In traditional herbal medicine, Indonesians generally use the purslane plant as an ingredient. This research aims to determine the antioxidant capacity and antibacterial properties of purslane plants using different solvents. The research methodology involved extracting purslane using different solvents, such as methanol, herbal ethyl, acetate, and hexane. Antioxidant testing used the DPPH method and compared the results with those obtained from vitamin C. This experiment employed the phytochemical diffusion method. Variations in solvent polarity may explain the differences between the results of phytochemical screening tests for various fractions and the results obtained from in silico toxicity tests using the Protox II and PKCsm methods. IC_{50} methanol fraction had an antioxidant activity of 96.12 ± 0.02, the ethyl acetate fraction 56.78 ± 0.04, the N-hexane fraction 456.72 ± 0.467, and Vitamin C 10.67 ± 0.02. The most effective against *E. Colli* is the ethyl acetate fraction with an inhibition zone value of 14.00 ± 0.10. Against *Pseudomonas aeruginosa* the ethyl acetate fraction was 17.77 ± 0.15, against *Salmonella typi* the N – Hexane fraction 15.24 ± 0.71, against *Bacillus subtilis* ethyl acetate fraction 14.77 ± 0.01, against *Streptococcus pneumonia* the N – fraction Hexane 14.08 ± 0.36 and against *Streptococcus aureus* ethyl acetate fraction 16.77 ± 0.17. The in silico toxicity prediction results classified the lutein component as a highly toxic component. However, not all purslane components show hepatotoxicity. Conclusion: The study found that the purslane herb fraction showed antioxidant properties, inhibited the growth of gram-negative and gram-positive bacteria, did not show hepatotoxicity, and had significant toxicity to the lutein component.



Keywords: Antibacterial, antioxidant, fractionation, Purslane herb

Introduction

The utilization of natural components as natural remedies derived from nature is an emerging subject of alternative medicine in Indonesia [1]. Back to Nature is a commonly chosen alternative by many individuals due to its healing capabilities and beneficial therapeutic results, which help reduce negative side effects for patients [2].

Purslane (*Portulaca oleracea* L.) can be utilized both as a culinary delicacy and as a therapeutic component. Purslane has been shown to contain high levels of omega-3 fatty acids, alkaloids, saponins, tannins, flavonoids, steroids, triterpenoids, and glycosides, according to experts at the University of Texas in San Antonio. Meanwhile, Spyridon Petropoulos and other researchers examined the molecular composition of the main active molecule found in *Portulaca oleracea* [3]. The chemicals in question are: a) lutein ($C_{40}H_{56}O_2$), b) α-tocopherol ($C_{29}H_{50}O_2$), c) β-carotene ($C_{40}H_{56}$), and d) ascorbic acid. The chemical formula C_6H_8OI represents a compound. Scientists have additionally recorded that purslane is abundant in melatonin, an antioxidant that suppresses cancer growth at a rate 10–20 times greater than that of other fruits or vegetables [4].

Portulaca oleracea studies have investigated the inhibitory effects of ethanol solvents on the proliferation of various bacterial strains, including *Pseudomonas aeruginosa, Salmonella typhimurium, Proteus mirabilis, Klebsiella pneumoniae,* and *Enterobacter aerogenes* [5]. This plant possesses the capacity to treat infectious illnesses while reducing numerous negative effects typically linked to synthetic antimicrobials an effective strategy to combat long-term antibiotic resistance is the

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advancement of natural medicine as a means to suppress bacterial growth [6].

Administration of dosages of 25, 50, and 100 mg/kg leads to notable alterations in the levels of non-enzyme antioxidants and enzymes in liver tissue, resulting in a considerable reduction in lipid peroxidation. Antioxidants are compounds that have the ability to counteract the detrimental impact of free radicals, which can lead to oxidative injury to skin cells and accelerate the aging process [7].

Prior research has conducted phytochemical extraction of *P. oleracea* using either ethanol or methanol (MeOH). Ethanol and methanol solvents are polar and have the ability to preferentially extract polar compounds [8], [9]. Solvents with partial polarity, such as ethyl acetate (EtOAc), are the only ones capable of extracting compounds with partial polarity. N-hexane, a non-polar solvent, exhibits the ability to exclusively extract non-polar compounds [10]. Hence, it is crucial to employ a diverse range of solvents capable of extracting molecules belonging to three distinct categories: polar, semi-polar, and nonpolar [2], [11]. Seldom do we employ other solvents, such as ethyl acetate (EtOAc) and n-hexane, for extraction. In this study, we performed extensive experimentation with disks in order to arrive at detailed findings regarding the minimal bacterial concentration. The objective of the study is to investigate the impact of n-hexane, EtOAc, and MeOH on the antioxidative and antibacterial capabilities of *Escherichia coli, Pseudomonas aeruginosa, Salmonella typhi, Bacillus subtilis, Streptococcus pneumonia*, and *Staphylococcus aureus*. The antioxidant activity of our test was measured using the DPPH method, with vitamin C serving as the benchmark for comparison [2], [12], [13], [14]

Materials and Methods

Materials

The plant in question is Purslane Herb (*Portulaca oleracea* L.). We utilize the entire plant, including all its parts. The samples were collected in Jl. Binjai Binjai City, North Sumatra. We conducted the sampling in January 2024. The Medannese Herbarium, Faculty of Mathematics, Natural Sciences, University of North Sumatra identified the purslane herb samples with Number 00123/UNJ.1.2.8.2/PPM/2024.

Ethical statement

The research was carried out at the Laboratory of the Faculty of Pharmacy, Universitas Sumatera Utara. After being thoroughly cleaned to remove any impurities, the purslane plant is dried in a drying cabinet at a temperature of 40–50°C until it reaches a state of dryness. The purslane herb was macerated using N-hexane, ethyl acetate, and methanol. The Faculty of Pharmacy, Universitas Sumatera Utara Laboratory analysed the purslane herb's phytochemical profile.

Materials

The research employs laboratory glassware, rotary evaporators, ovens, incubators, and UV-Vis spectrophotometry (Shidmazu, 1700). The study utilized purslane herb samples and solvents such as N-Hexan, ethyl acetate, and methanol. Additionally, NaOH 2N and 1,1-diphenyl-2-picryl-hydrazyl (DPPH) were also used. Gram-positive bacteria, such as *Staphylococcus aureus, Bacillus subtilis, and Streptococcus pneumoniae*, as well as gram-negative bacteria including *Escherichia coli, Pseudomonas aeruginosa, and Salmonella typhi*, were used in the experiment. The dragendroff recombinant, 10% HCL, and nutrient agar were also utilized.

Maceration, Fractionation and Phytochemical Screening of MeOH Extracts of *P. oleracea*

We obtained two samples of *P. oleracea* dried powder, each weighing 1 kg. We used 3L of methanol solvent to extract the samples for 24 hours. After extraction, we filtered the samples but forgot to collect the *P. oleracea* methanol extract. The resulting extract was fractionated by employing N-hexane solvent six times over two hours, with each repeat utilizing 500 mL of N-hexane. Upon finishing, we introduced 300 mL of water to *the P. oleracea* methanol fraction to enhance its polarity. Subsequently, we included 400 mL of ethyl acetate into the fraction for 2 hours. This process was reiterated up to a maximum of ten times. Upon the completion of the fractionation procedure, all fractions were consolidated into a single container and concentrated using a rotary evaporator to get the methanol fraction, ethyl acetate fraction, and N-hexane fraction of *P. oleracea* [3], [4]. Phytochemical screening 500 mg of methanol, ethyl acetate, and N-hexane fraction of *P. oleracea* were combined in a test tube. Then, 3 ml of methanol was added and mixed thoroughly. Phytochemical screening aims to ascertain the presence and quantity of secondary metabolite chemicals in each fraction [2].

Screening of antibacterial activity

Diffusion tests conduct antibacterial screening to ascertain the antibacterial activity using the diffusion test allows the researcher to predict the concentration of the antibacterial agent being studied (Kirby-Bauer methods) [15], [16].

Medium Preparation

The medium employed was nutrient agar (NA). A total of 10 grams of NA were added to a 1 liter Erlenmeyer flask after pouring in 500 millilitres of distilled water. The mixture was agitated and heated on a hotplate, then sterilized in an autoclave at a temperature of 121°C [17].

Bacterial strains

The bacterial strains utilized in the study were *Staphylococcus aureus* (FNCC-0027) and *B. subtilis* (FNCC-0035) for gram-positive bacteria, and *E. coli* (FNCC-0145) and *P. aeruginosa* (ATCC 2467) for gram-negative bacteria. Both Gram-positive and Gram-negative bacteria were acquired from Gadjah Mada University. The other bacterial strains utilized were *S. pneumoniae* (ATCC 70184) (gram-positive) and *S. typhi* (ATCC 14028), acquired from the Microbiology Laboratory of the Faculty of Pharmacy, Universitas Sumatera Utara.

Inoculum standardization preparation

Bacterial cultures were inoculated onto agar media and subjected to incubation at a temperature of 37°C for 18–24 hours. Subsequently, the bacterial culture was mixed with physiological saline solution (NaCl) to reduce its concentration. The turbidity of the mixture was adjusted to match a McFarland turbidity of 0.5%, equivalent to a concentration of 1 x 108 CFU/ml [13], [18].

Activity test antibacterial activity with Disc Diffusion Method

First, a disc paper measuring 6 mm in size was prepared. Then, it was soaked with 100 µl of methanol fraction, ethyl acetate fraction, and N-hexane fraction of *P. oleracea* to achieve a concentration of 10.000 ppm. A positive control for the tetracycline test was also prepared with a concentration of 200 ppm. Subsequently, paper disks moistened with the test solution

were inserted into Petri dishes containing NA medium and incubated at 37°C for 24 hours. After 24 hours, the specimen was extracted from the incubator, and the transparent region was assessed by a calliper [19].

Preparation of Vitamin C Standard Solution

Ten mg of standard vitamin C was dissolved in 2 ml of ethanol and then diluted with distilled water to a final volume of 100 ml. The solution was further diluted to achieve concentrations of 50, 25, 15, and 5 parts per million (ppm) [20].

DPPH free-radical scavenging activity

The 1,1-diphenyl-2-picryl-hydrazyl (DPPH) technique was employed to conduct antioxidant testing. The fundamental basis of this test lies in the transformation of color, namely from purple to yellow, as a result of the mechanism of antioxidant response. The solutions were prepared by diluting the extract with 200, 100, 50, and 10 µg/ml concentrations in 2 ml of methanol. Then, 0.5 ml of DPPH (1 mM in methanol) was added. Allow the test solution to remain undisturbed for 30 minutes until it undergoes a reaction with DPPH. Utilize UV-Vis spectrophotometry to quantify the absorbance at a specific wavelength of 517 nm [1]. Calculate the percentage of inhibition by calculating the disparity in absorbance between the blank solution and the sample, employing the following formula:

$$\%\text{DPPH Free radical scavenging activity} = 1 - \{\frac{\text{the absorption of control} - \text{the absorption of sampel}}{\text{the absorption of control}}\}$$

In silico Tools

The hardware used in this research consists of a collection of HP Pavilion Gaming Laptops with 15-ec2xxx 64-bit specifications and a 1 TB hard disk, as well as software and operating system software. We used Windows 11, Pubchem, pK-CSM Tools, and Pro-Tox II databases. Search for structure-based and ligand-based compounds in sample molecules.[21], [22].

Preparation of Compound for in silico Toxicity Prediction

The generation of Canonical SMILES for each compound was performed using the Pubchem website (https://pubchem.ncbi.nlm.nih.gov/) [23].

Toxicity Prediction of Compound with pK-CSM Tools

To predict compound toxicity, input the Canonical SMILES and click the ADMET option. This will allow us to use the pK-CSM Tools available at http://biosig.unimelb.edu.au/pkcsm/prediction. These tools provide the distribution of absorption analysis results, including VDss (volume of distribution at steady state), fraction unbound, BBB permeability (blood-brain barrier permeability), and CNS permeability (central nervous system permeability). Metabolism and toxicityThe generation of Canonical SMILES for each compound was performed using the Pubchem website (https://pubchem.ncbi.nlm.nih.gov/) [21], [24].

Toxicity Prediction of Compound with Pro-Tox II

To predict compound toxicity using Pro-Tox II, go to https://tox-new.charite.de/protox_II/. Click on "Tox Prediction" and enter the Canonical SMILES. Select all toxicity parameters and initiate the Tox-Prediction process. This will provide with the results of the compound's toxicity analysis, including LD50, hepatotoxicity, carcinogenicity, immunotoxicity, mutagenicity, cytotoxicity, AhR, AR, AR-LBD, aromatase, ER, ER-LBD, PPAR-Gamma, nrf2/ARE, HSE, MMP, phosphoprotein tumour suppressor, and ATAD 5 [21], [25].

Data Analysis

Data analysis using SPSS version 22

Results and Discussion

Phytochemical Screening

Phytochemical screening tests were conducted to assess and compare the secondary metabolite composition of the test materials. The outcomes of the secondary metabolite examination for each extract tested are displayed in Table 1.

Table (1): Phytochemical screening results of methanol extract, ethyl acetate fraction and N-hexane fraction of purslane (*Portulaca oleracea* L.) herb.

No	Parameters	Observation	MeOH Fraction	EtOAc Fraction	N – Hexane Fraction
1.	Alkaloids	Orange precipitate	+	+	+
2.	Phenolics	Purple	+	+	+
3.	Flavonoids	Orange-red colored solution	+	+	-
4.	Triterpenoid	Red-purple solution	-	+	+
5.	Steroids	Green colored solution	-	-	-
6.	Saponins	The foam does not disappear after the addition of concentrated HCl	+	+	+

Information :

(+) Contains coumpounds

(-) doesn't Contains coumpounds

Antioxidant Activity Test of Methanol, Ethyl Acetate, and N-Hexane Fractions of *Portulaca oleracea* L.

The DPPH method was employed to assess the antioxidant activity of the test sample, with the IC_{50} value used to quantify its antioxidant activity. The test results employed vitamin C as a benchmark or standard positive control. Antioxidant activity refers to the capacity of a substance or extract to hinder oxidation reactions, which the percentage of inhibition can quantify.

The antioxidant activity test findings of the test samples are displayed in Table 2 below. Figure 1 below provides a visual comparison of solvents.

Table (2): Antioxidant activity of each fraction of *P. oleracea*

Sample	IC_{50} (µg/mL)
MeOH Fraction	96.12 ± 0.02
EtOH Fraction	56.78 ± 0.04
N – Hexane Fraction	456.72 ± 0.467
Vitamin C	10.67 ± 0.02

Antibacterial Activity Test by the Disc Diffusion Method

The antibacterial activity of the *P. oleracea* extract was tested against *Escherichia coli, Pseudomonas aeruginosa, Salmonella typi, Bacillus subtilis, Streptococcus pneumonia, and Staphylococcus aureus*. Tetracycline was used as a positive control, and a DMSO solution was used for comparison. The antimicrobial testing findings of the *P. oleracea* extract are displayed in Table 3

Table (3): Antibacterial activity in each fraction *P. oleracea*

Sampel	*Escherichia coli* (mm)	*Pseudomonas aeruginosa* (mm)	*Salmonella typi* (mm)	*Bacillus subtilis* (mm)	*Streptococcus pneumonia* (mm)	*Staphylococcus aureus* (mm)
MeOH Fraction	7.70 ± 0,10	7.77 ± 0,15	8.87 ± 0,06	7.03 ± 0,47	9.47± 0.31	9.83 ± 0.06
EtOH Fraction	14.00 ± 0,10	17,77 ± 0,15	12,77 ± 0,55	14,77 ± 0,01	10,77 ± 0,25	16,77 ± 0,17
N – Hexane Fraction	10,24±0,59	11.03±0,62	15,24±0,71	10,83±0,40	14,08±0,36	11,17±0,88
Tetracyclin	24,67±0,29	22,67±0,48	23,84±0,35	21,54±1,07	21,28±0,89	24,10±0,62
DMSO	-	-	-	-	-	-

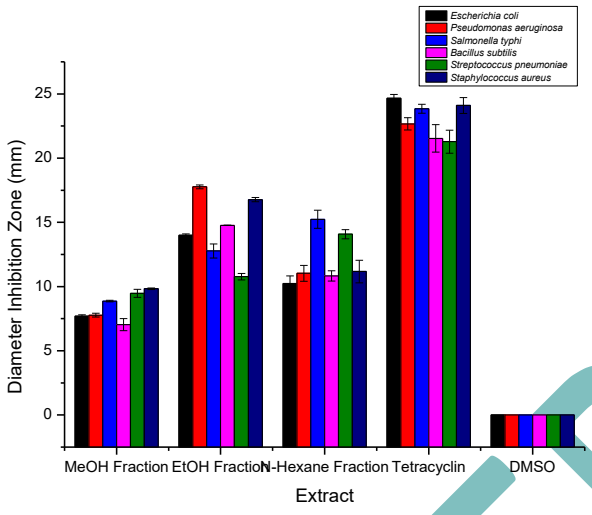

Figure (1): The relationship between bacterial inhibition and the test sample.

The evaluation of antibacterial activity is conducted using the disc-paper method. The test sample will permeate the disc paper, which will be inserted into a culture medium colonized by bacteria and incubated for 18-24 hours. Subsequently, the transparent area will be observed and quantified. The formation of a clean zone demonstrates the test sample's capacity to prevent the growth of bacteria [26]. This study conducted tests on the test samples of methanol extract, ethyl acetate fraction, and N-Hexane to assess their ability to hinder the growth of both gram-negative and gram-positive bacteria. The test results indicate a disparity between the inhibition of each test sample and the bacteria examined.

Toxicity Prediction of Compounds

Isocitric, citric acid, lutein, α-tocopherol, β-carotene, ascorbic acid, and glutathione are a group of bioactive compounds found in *P. oleracea*. Asra's 2020 research indicated that the average betacyanin levels in the red beetroot extract (*Beta vulgaris* L.) were 98.6474% ± 0.584080 [24]. The active compound utilized in this study is an in silico toxicity test. We performed toxicity profile testing and ADME analysis using the HP Pavilion Gaming Laptop 15-ec2xxx 64-bit laptop equipment, the pKCSM online tool application available at http://biosig.unimelb.edu.au/pkcsm/, and the Protox application online tool accessible at https://tox-new.charite.de/protox_II/. The results of ADME and toxicity profiling are presented in Tables 4-6.

Table (4): Profile ADME

Property	Model Name	Predicted Value							Unit
		Isocitric	Citric acid	Lutein	α-tocopherol	β-carotene	ascorbic acid	glutathione	
Absorption	Water Solubility	-1.832	-1.423	-6.822	-6.901	-7.39	-1.556	-2.892	Numeric (log mol/L)
Distribution	VDss (human)	-0.712	-0.418	-0.23	0.709	0.266	0.218	-0.377	Numeric (log L/kg)
Metabolism	CYP2D6 Substrate	No	No	No	No	No	No	No	Categorical (Yes/No)
Excretion	Total Clearance	0.892	0.895	0.924	0.794	1.061	0.631	0.308	Numeric (log ml/min/kg)

Table (5): Prediction toxicity (pKCSM)

Property	Model Name	Predicted Value							Unit
		Isocitric	Citric acid	Lutein	α-tocopherol	β-carotene	ascorbic acid	glutathione	
Toxicity	AMES toxicity	No	No	No	No	Yes	No	Yes	Numeric (log ml/min/kg)
Toxicity	Max. tolerated dose (human)	0.97	0.749	-1.068	0.775	-0.379	1.598	1.104	Categorical (Yes/No)
Toxicity	hERG I inhibitor	No	No	No	No	No	No	No	Categorical (Yes/No)
Toxicity	hERG II inhibitor	No	No	Yes	Yes	Yes	No	No	Numeric (log mg/kg/day)

Toxicity	Oral Rat Acute Toxicity (LD50)	2.288	2.148	3.491	2.072	2.073	1.063	2.468	Categorical (Yes/No)
Toxicity	Oral Rat Chronic Toxicity (LOAEL)	3.564	3.698	2.572	1.987	0.65	3.186	2.919	Categorical (Yes/No)
Toxicity	Hepatotoxicity	No	No	No	No	No	No	No	Numeric (mol/kg)
Toxicity	Skin Sensitisation	No	No	No	No	No	No	No	Numeric (log mg/kg_bw/day)
Toxicity	T.Pyriformis toxicity	0.285	0.285	0.335	1.017	0.326	0.285	0.285	Categorical (Yes/No)
Toxicity	Minnow toxicity	4.245	4.251	-2.213	-3.324	-4.028	4.386	4.569	Categorical (Yes/No)

Table (6): Toxicity class (protox online)

No.	Parameters	Isocitric	Citric acid	Lutein	α-tocopherol	β-carotene	ascorbic acid	glutathione
1.	Predicted LD 50	80 mg/kg	80 mg/kg	10 mg/kg	5000 mg/kg	1510 mg/kg	3367 mg/kg	5000 mg/kg
2.	Predicted toxicity class	Class 3	Class 3	Class 2	Class 5	Class 4	Class 5	Class 5
3.	Average similarity	100%	100%	73.04%	82.25%	83.45%	100%	100%
4.	Prediction accuracy	100%	100%	69.26%	70.97%	70.97%	100%	100%

The phytochemical screening results show that different levels of secondary metabolites are present in the methanol, ethyl acetate, and n-hexane fractions of *Portulaca oleracea* L. These variations are probably caused by the different solvent polarity utilised during the extraction procedure. In line with earlier research by Karmakar et al. (2021) which verified that P. oleracea is abundant in bioactive compounds, the presence of active compounds like alkaloids, phenols, flavonoids, triterpenoids, and saponins in each fraction suggests that purslane has a diverse chemical complexity [27]. The biological activity of each fraction is directly implied by the difference in metabolite content. The choice of solvents becomes extremely important in this situation since it dictates the kinds of molecules that are extracted, which in turn influences the extract's pharmacological efficacy [28].

The ethyl acetate fraction has the greatest potential for scavenging free radicals, according to the produced antioxidant activity. Its capacity to extract semi-polar substances like flavonoids and polyphenols—which are known to contain active hydroxyl groups that operate as electron or hydrogen donors and are therefore useful in the radical scavenging mechanism—explains this [29]. These outcomes corroborate those of Tleubayeva et al. (2022), who found that the ethyl acetate fraction of P. oleracea exhibited strong antioxidant activity [7]. The measured IC_{50} value shows the promising potential of this fraction to be developed as a candidate for natural antioxidants, even though it is not as strong as that of vitamin C. This is significant since the food and pharmaceutical industries frequently experience long-term harmful side effects from the usage of synthetic antioxidants. As a result, the findings of this study enhance the natural antioxidant sources from indigenous Indonesian plants that have not yet been thoroughly investigated [30]. However, each fraction's antibacterial activity demonstrates different capacities to inhibit both Gram-positive and Gram-negative bacteria. The inclusion of flavonoids and alkaloids, which have been shown to alter bacterial cell membrane structures and inhibit vital enzymes required for microbial development, is probably why the ethyl acetate fraction once again shown the highest level of activity [31], [32]. These findings are consistent with a research by Thu et al. (2023) that demonstrated a substantial inhibitory zone against *Staphylococcus aureus* in the purslane ethyl acetate fraction [33]. The intricacy of the bacterial species' cell wall architecture is also reflected in the variations in susceptibility to the extract; Gram-negative bacteria are typically more resistant because of the hydrophobic lipopolysaccharide layer they have [34]. Triterpenoids and other non-polar substances that can integrate into the lipid membrane and cause cell leakage are responsible for the n-hexane fraction's highest activity against *Streptococcus pneumoniae* and *Salmonella typhi* [35].

This conclusion is significant not only because it shows that purslane extract has antibacterial properties, but also because it shows that the plant's biological activity spectrum is more fully represented by the polarity-based tiered extraction method. This method makes a significant methodological contribution to the study of Indonesian herbal plants because it is quite uncommon in local research. Additionally, in silico toxicity predictions offer a preliminary understanding of the safety of the bioactive substances found in P. oleracea [21]. Other substances including ascorbic acid, glutathione, and α-tocopherol demonstrated a good safety profile, including no signs of hepatotoxicity, despite lutein's high toxicity potential (low LD_{50} and toxicity class. Because they address worries about the possible negative effects of using natural chemicals as therapeutic agents over an extended period of time, these findings are significant. This study effectively overcomes the drawbacks of in vivo toxicology testing, which frequently demand a large investment of time and resources, by employing a computational toxicology technique. These findings may eventually be used as a foundation for choosing and altering the chemical structure of substances that are highly active but not harmful [36], [37], [38].

All things considered, this study significantly advances the science of natural product pharmacy, especially in the area of developing potential antibacterial and antioxidant compounds from indigenous Indonesian plants. In the age of contemporary drug discovery, the combination of phytochemical screening, bioactivity testing, and in silico toxicity prediction represents a multidisciplinary approach that is becoming more and more pertinent [39] . The lack of time-kill and minimum inhibitory

concentration (MIC) experiments to guarantee antibacterial activity, as well as the absence of in vivo studies to assess pharmacological and toxicological effects directly, are among of the study's shortcomings. Furthermore, despite its efficiency, the in silico method is still not perfect for forecasting intricate interactions in actual biological systems. It is important to carefully analyse the results and refrain from making direct generalisations for therapeutic applications in light of these limitations [22], [40]. These results still offer a solid basis for more research, whether it takes the shape of sophisticated in vivo studies, the separation of pure substances, or the creation of medication formulations utilising *Portulaca oleracea* L. bioactive fractions.

Conclusion

The research findings demonstrated that *Portulaca oleracea* L, when subjected to four solvents of varying polarity, had the capacity to effectively hinder the growth of both gram-positive and gram-negative bacteria. The findings suggest that the increasing understanding of the relationship between learning and herbal formulations including *Portulaca oleracea* L holds significant promise for the treatment of illnesses resulting from oxidative stress and bacterial infections. Additionally, they can be utilized in the production of pharmaceuticals or dietary supplements for the chemical sector in the field of medicine.

Ethics approval and consent to participate

Not applicable

Consent for publication

Not applicable

Availability of data and materials

Not applicable

Author's contribution

The authors confirm contribution to the paper as follows: **N.N.** (Nasri Nasri) contributed to the study conception and design, data analysis and validation, as well as manuscript preparation. **V.E.K.** (Vera Estefania Kaban) was responsible for the study conception and design, as well as theoretical calculations and modeling. **M.F.L.** (Muhammad Fauzan Lubis) contributed to data collection and analysis, and manuscript preparation. **S.E.N.** (Sony Eka Nugraha) assisted with data analysis and validation and contributed to manuscript editing. **W.W.** (Wahyudi Wahyudi) participated in data analysis and validation, and manuscript review. **C.G.T.** (Chyntia Glori Tania) contributed to data collection and analysis, and manuscript review. **N.S.** (Nurul Suci) was involved in data analysis and validation, and manuscript editing. **Z.R.** (Zulmai Rani) contributed to data collection and manuscript editing. All authors reviewed the results and approved the final version of the manuscript.

Funding

This study was not funded

Conflicts of interest

The authors declare no conflict of interest.

Acknowledgements

The authors would like to express their gratitude to the institutions involved in this research and the preparation of this manuscript. Special thanks to Universitas Sumatera Utara (USU), Universitas Prima Indonesia (UNPRI), Universitas Islam Negeri Sumatera Utara (UINSU), and Universitas Muslim Nusantara Al-Washliyah (UMN Al-Washliyah) for their invaluable support and collaboration throughout the study.

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