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Research Article

The Efficacy of Flavonoids from Potato Peels (*Solanum Tuberosum*) Extracts Against *Listeria monocytogenes* Causing Listeriosis

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I N F O

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A B S T R A C T

Background: Listeriosis, caused by *Listeria monocytogenes*, remains a serious food-borne threat. Continuous monitoring and alternative antimicrobials are needed to curb emerging virulent, drug-resistant strains.

Objective: To assess the prevalence of *L. monocytogenes* in raw milk and evaluate the in-vitro anti-*Listeria* efficacy of flavonoid-rich potato-peel extracts.

Methods: Sixteen raw-milk samples were cultured; isolates were confirmed as L. monocytogenes. Potato peels were extracted with 80 % methanol and other solvents. Total flavonoid content and individual compounds were quantified (HPLC). Minimum inhibitory concentrations (MICs) of the extracts were determined against the isolates and compared with gentamicin and amikacin.

Results: L. monocytogenes was detected in 6/16 samples (37.5 %). The 80 % methanolic extract gave the highest yield and total flavonoids. Quercetin was the dominant flavonoid, followed by hesperidin, rutin, and naringin. The methanolic extract inhibited all isolates with MICs of 8–64 µg ml^{-1}, exhibiting stronger activity than gentamicin or amikacin.

Conclusion: Potato-peel flavonoids, especially quercetin-rich methanolic extracts, show potent in-vitro activity against L. monocytogenes, highlighting their promise as natural antimicrobials or adjunct treatments for listeriosis.

Keywords: *Listeria monocytogenes*, flavonoids, foodborne infection, potato peels

Introduction

Most *L. monocytogenes* infections in humans occur during pregnancy or when there is immunosuppression due to sickness or medication.[1, 2] There is mounting evidence that links the food-borne spread of *L. monocytogenes* to a significant number of human listeriosis cases.[3, 4] Contrary to the majority of foodborne infections, which mostly cause gastrointestinal disorders, invasive illnesses caused by *L. monocytogenes* include meningitis, sepsis, chorioamnionitis, and stillbirth. It is second most common cause of fatality among neonates and infants.[5]

The genus *Listeria*, which has seven species, is found throughout nature. It is a common intracellular Gram-positive non-sporulating pathogenic bacterium. *Listeria* has been found in slaughterhouse waste, sewage, water, animal feed, fresh and frozen meat, poultry, and human excrement. These organisms have the potential to become endemic in environments where food is processed.[6, 7] Several domestic and wild animals, as well as people, are afflicted by L. monocytogenes.[5, 8] Consumption of tainted food causes infection in the vast majority of human cases.[3, 6]

The potato (*Solanum tuberosum*) is one of the vegetables most widely consumed.[9] The primary byproduct of the potato processing industry is potato peel waste, which has the potential to include a variety of functional and bioactive substances, including pigments, dietary fiber, vitamins, and minerals, in addition to antioxidants.[10] Potato peels, a waste byproduct of potato processing, have shown antioxidant activity in numerous in vitro test methods;[11] additionally, the aqueous extract of potato peels is high in phenolic acids, such as hydroxycinnamic acids and flavonoids, which have a high antioxidant activity[12] and give therapeutic advantages, including erythrocyte protection, without causing mutagenesis[11, 13] Consequently, the goal of this study was to assess the flavonoids in potato peel extracts as antibacterial substances against *L. monocytogenes* isolated from raw milk.

Materials and Methods

Preparation of Potato Peel Extract

Potato skins were cleaned, dried at 40°C, then milled into a fine powder. The sample was filtered using Whatman filter paper after being extracted with 100 ml of each of the organic solvents methanol, ethanol, hexane, and petroleum ether overnight at room temperature in a shaker. The residues were extracted again under the same conditions. In the rotary evaporator below, the mixed filtrates were heated to 35°C. After organic solvents were removed from the extracts, the extract yield was calculated, and the extracts were kept at -20°C.[14]

Ethical Approval

Ethical approval was not required for this study as it did not involve human participants or animal subjects. All work was limited to in-vitro experiments using food and plant-derived materials.

Total Flavonoids Determination

To the extract solution, a 0.5 mL aliquot of a 2% AlCl3 ethanolic solution was added. The absorbance at 420nm was measured after an hour at room temperature. A golden tinge suggested flavonoids were present. The sample extracts were evaluated at 0.1mg ml-1 of the final concentration of the extract. The following equation was used to calculate the total flavonoid content expressed as quercetin equivalent (QE) based on the calibration curve (15). Y=0.0255x: where x: absorbance and y: concentration.

HPLC

The chromatographic investigation employed a 4.6 mm ID x 250 mm Zorbax SB-C18 column and a Waters HPLC 2690 equipped with a diode array detector, automated injector, and automatic degassing system. A sample volume of 10 microliters was injected. The circumstances of the analysis were as indicated with[16] a flow rate of 1 ml/min of HPLC grade methanol and 1% (v/v) formic acid in water (A). The absorbance was measured between 254 and 324 nm in wavelength.

Isolation of *Listeria* from raw milk

At random, sixteen samples of raw milk from various locations around Baghdad city's supermarkets were taken (The sample size of sixteen raw milk samples was selected based on feasibility and availability of resources, in line with similar exploratory studies focused on the isolation and preliminary screening of *Listeria monocytogenes* in food products). Oxford agar, a specialized medium for isolating *L. monocytogenes*, was cultured on samples of milk, and the cultures were kept at 37°C for 48 hours. Based on morphological examination, staining, and biochemical analysis, isolated bacterial cultures (*L. monocytogenes*) have been identified. After incubation, colonies that appeared were most likely *L. monocytogenes*, thus, they were subcultured onto nutrient agar for further identification.[13]

Antibacterial activity of flavonoids against *L. monocytogenes*

By determining the MIC for flavonoids, gentamicin, and amikacin as follows,[18] the microdilution test in microplates was performed to assess the effect of these drugs on *L. monocytogenes* isolates. Bacterial inoculum from an overnight-growing culture was compared to a 0.5 McFarland turbidity standard. Each well received 80ml of various flavo-

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noid concentrations, 100 ml of Mueller-Hinton broth, 20 ml of microbial inoculum, and gentamicin or amikacin (2-1024 g/ml). At 37°C, the microtiter plates were incubated for a full day. The MIC was established as the lowest dose at which the organism failed to demonstrate discernible growth.

Results and Discussion

Determination of Total Flavonoids in Potato peels extract

Figure (1) presents the qualities of potato peel extracts. In comparison to other extracts, the methanol extract (80%) had a greater yield and total Flavonoids. The lowest characteristics were found in the hexane extract, though. 80% methanol extract and ethanol extract both contained comparable amounts of total flavonoids, and both extracts had higher levels of total flavonoids than other extracts. Methanol and ethanol were shown to be the most effective solvents, yielding considerable quantities of yield due to their greater polarity and superior solubility from plant components.[16] Total flavonoid extract was frequently less plentiful in water and more abundant in methanol, with ethanol in potato peel extract coming in second.[17]

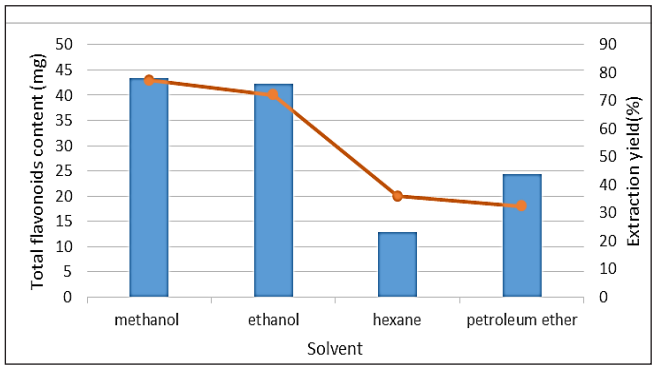

Figure 1.Extraction yield and total flavonoids content in potato peels with different solvents

HPLC analysis

According to HPLC analysis the flavonoid component quercetin was the most prevalent in the methanol extract of potato peels. Compared to the traditional sources of flavonoids, hesperidin, rutin, and naringin come in second. According to Hsieh, Y et al (2016)[18] the flavonoid components quercetin, hesperidin, naringin, and rutin were present in the water extract. While Chu, Y. et al (2000) (19) revealed that the flavonoids in potato peels are made up of myricetin, quercetin, and kaempferol.

Isolation of *Listeria* from raw milk

Six *L. monocytogenes* isolates were produced after 16 samples of raw milk were cultured on the selective medium Oxford agar. *L. monocytogenes* hydrolyzes esculetin into esculetin, which subsequently interacts with ferric ammonium citrate to form a precipitate and a reaction in this solid media. The presence of gram-positive rods is shown by staining. Listeriosis, a zoonotic illness, is often spread by the consumption of infected food. *L. monocytogenes* was found in two samples, including raw milk (26%), ice cream (15%), and yogurt (14%). It was shown that raw milk was the main source of L. monocytogenes. There was *Listeria* contamination in about 25% of the raw milk. This is a result of poor hygiene, contaminated environments, and milk handlers.[5] Raw beef had the highest level of *Listeria* species contamination, followed by raw milk and liquid whole eggs.[3]

Antibacterial activity of flavonoids against *L. monocytogenes*

By evaluating MICs, the effectiveness of extracted flavonoids and widely used antibiotics gentamicin and amikacin against *L. monocytogenes* was determined. Amikacin was shown to be more effective than gentamicin, and most *L. monocytogenes* isolates showed only mild resistance to those two antibiotics. For *L. monocytogenes* isolates, however, the isolated flavonoids from potato peels demonstrated a higher amount of inhibition, with MIC values reaching 8–64 µg/ml (figure 3).

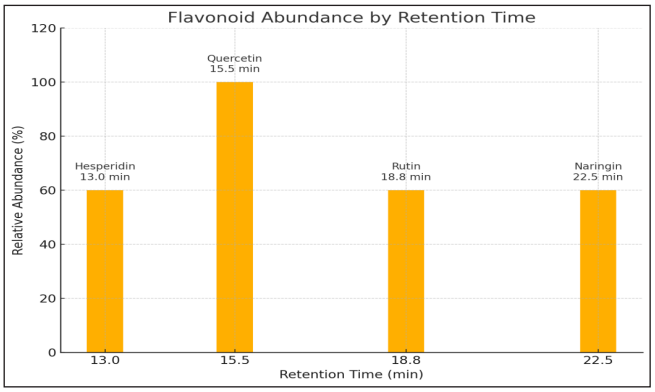

Figure 2.HPLC analysis for total flavonoids content extracted from potato peels Isolation of *Listeria* from raw milk

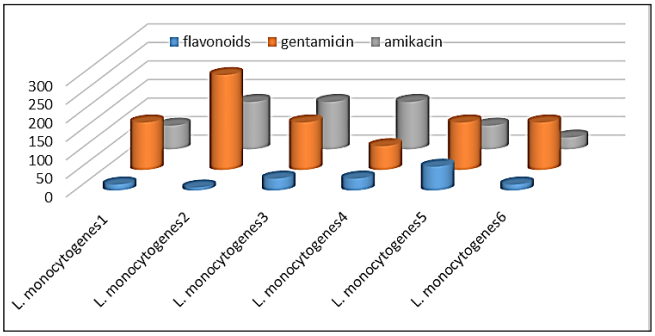

Figure 3.Minimum inhibitory concentrations of antibiotics and flavonoids against *L. monocytogenes*

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L. monocytogenes strains were more likely to exhibit resistance to the antibiotics cephalosporins, aminoglycosides, and tetracyclines. The intermediate resistance for clarithromycin, gentamicin, and amikacin revealed may imply that *L. monocytogenes* is gradually accumulating antibiotic resistance.[6]

Antimicrobial resistance in *L. monocytogenes* is influenced by some variables, particularly in the food industry. Additionally, exposure to the stress associated with food processing may promote the emergence of antibiotic resistance that is relevant to clinical settings.[5] The Rajasthan region's raw milk contains multidrug-resistant *L. monocytogenes* strains, which poses a risk to the public's health and highlights the need for more consumer education and the adoption of tougher food safety laws at all stages of milk production.[20]

Conclusion

Quercetin is the most prevalent flavonoid constituent in the potato peel methanol extract, followed by hesperidin, rutin, and naringin. In comparison to the most commonly used antibiotics like gentamicin and amikacin, the extracted flavonoids from potato peels demonstrated a higher level of inhibition for *L. monocytogenes* isolates with MIC values reaching 8-64µg/ml. Total flavonoids may therefore have a significant role in the treatment of *Listeria monocytogenes*, a dangerous invasive disease.

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Declaration of Generative AI and AI-Assisted Technologies in the Writing Process: None

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