

Acknowledgments

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Unveiling the Antimicrobial Properties of Raspberry Leaves Against Multidrug-Resistant *Klebsiella pneumoniae*

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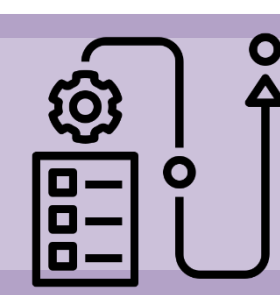
Introduction

- *Klebsiella pneumoniae*, a major contributor to nosocomial infections, accounts for 10% of hospital-acquired infections and is classified as a critical and high-priority antibiotic-resistant bacterium by the WHO.
- Infections stemming from *K. pneumoniae* are often severe and life-threatening due to high levels of antibiotic resistance, resulting in adverse outcomes.
- Consequently, there is an urgent need for new drugs effective against *K. pneumoniae* infections. One promising approach involves exploring the antibacterial potential of natural resources, specifically byproducts rich in bioactive compounds, notably phenolic compounds.



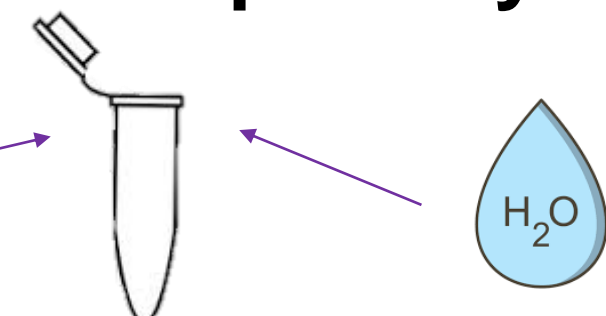
Aims

- Assess the antimicrobial, phytochemical, and antioxidant activities of aqueous extracts from raspberry leaves against multidrug-resistant *K. pneumoniae* strains isolated from clinical wound infections



Methods

- Preparation the raspberry's extracts

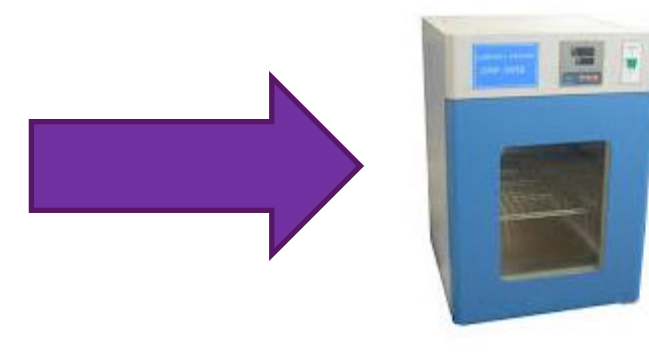


Final concentration of 1 mg/ml

1 mg

1ml

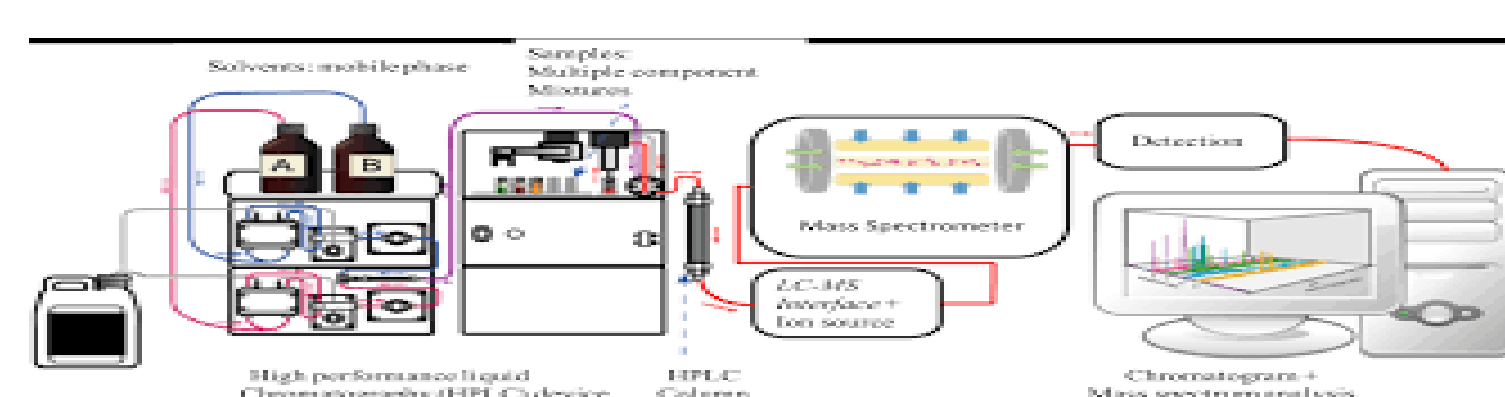
- 24 hours bacteria incubation in BHI



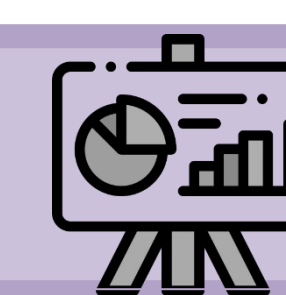
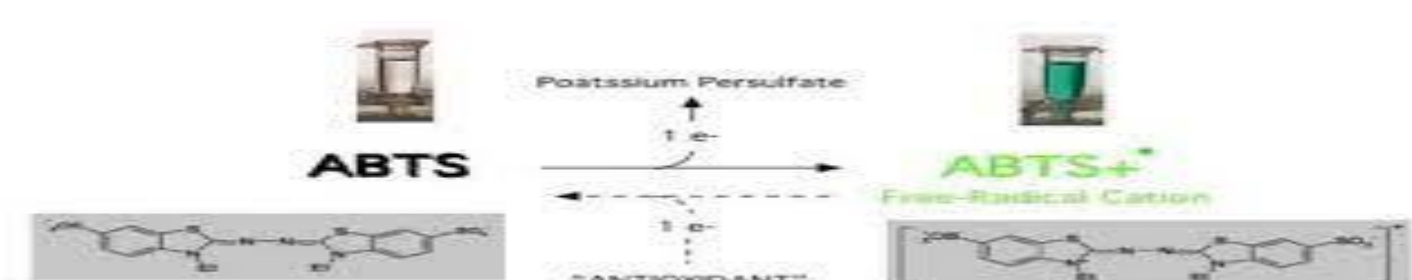
Incubation 24 hours at 37°C

- Antimicrobial activity

- Quantification of phenolic compounds by LC-MS



- Antioxidant activity using the ABTS method

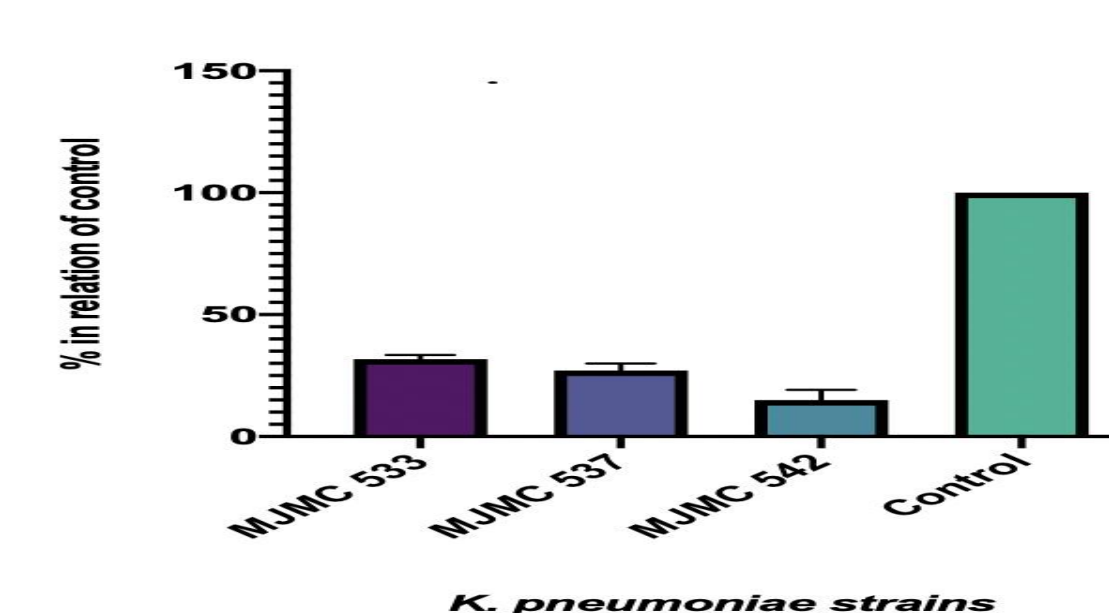


Results

- **Table 1.** Retention time (Rt), wavelengths of maximum absorption in the visible region (λ_{max}), mass spectral data, identification and quantification (mg/g of extract) of phenolic compounds in raspberry aerial parts.

Peak	Rt (min)	λ_{max} (nm)	[M-H] ⁻ m/z	MS ² (m/z)	Identification	Decoction	Infusion	Maceration	UAE
1	4.60	324	353	1911(00), 1795(5), 135(10)	3-O-Caffeoylquinic acid ¹	5.38±0.05 ^a	7±1 ^a	8.0±0.3 ^a	3.9±0.2 ^a
2	5.09	322	707	467(2), 353(8), 300(8), 191(15)	Caffeoylquinic acid dimer ¹	4.02±0.05 ^a	3.48±0.04 ^b	nd	2.6±0.2 ^a
3	6.01	310	341	179(8), 149(5), 135(10)	Caffeic acid hexoide ²	1.76±0.02 ^a	1.466±0.002 ^a	1.26±0.02 ^a	1.1±0.1 ^a
4	6.67	322	707	467(2), 353(8), 300(8), 191(15)	Caffeoylquinic acid dimer ¹	40.7±0.4 ^a	43.3±0.3 ^a	46.0±0.4 ^a	46.0±0.4 ^a
5	7.77	323	353	1911(00), 1798(5), 135(10)	5-O-Caffeoylquinic acid ¹	3.7±0.1 ^a	4.1±0.04 ^b	nd	nd
6	8.95	283	863	711(2), 573(1), 453(1), 413(1), 289(6)	Procyanidin trimer ³	5.8±0.1 ^b	6.2±0.1 ^b	4.1±0.1 ^a	8.8±0.1 ^a
7	9.91	282	863	711(2), 573(1), 453(1), 413(1), 289(6)	Procyanidin trimer ³	7.7±0.1 ^a	6.2±0.1 ^b	4.5±0.1 ^a	7.3±0.1 ^a
8	12.23	282	1153	865(3), 577(1), 575(1), 561(7), 289(13)	Procyanidin tetramer ³	0.73±0.00 ^a	0.92±0.01 ^a	0.64±0.00 ^a	nd
9	15.51	281	1153	865(3), 577(1), 575(1), 561(7), 289(10)	Procyanidin tetramer ³	5.9±0.03 ^a	7.57±0.02 ^a	5.2±0.1 ^a	nd
10	16.71	345	609	301(10)	Quercetin-3-O-rutinoside ⁴	2.64±0.02 ^a	2.45±0.02 ^a	2.73±0.02 ^a	2.47±0.01 ^a
11	17.79	327	463	301(10)	Quercetin-3-O-glucoside ⁴	2.67±0.01 ^a	2.10±0.02 ^a	2.96±0.02 ^a	2.90±0.02 ^a
12	19.35	319	515	353(10), 191(11), 179(8)	3,5-O-Dicaffeoylquinic acid ⁵	0.72±0.04 ^a	0.575±0.004 ^a	0.669±0.004 ^a	0.332±0.004 ^a
13	19.79	340	593	285(10)	Luteolin di-6,8-C-hexoside ⁶	4.87±0.05 ^a	4.4±0.1 ^a	4.9±0.1 ^a	5.0±0.2 ^a
14	21.03	334	447	285(10)	Luteolin 6-C-glucoside ⁶	3.5±0.1 ^b	3.07±0.05 ^a	4.4±0.1 ^a	4.0±0.2 ^a
					TPA	56.3±0.1 ^a	60±1 ^a	56.0±0.1 ^a	54±1 ^a
					TP	20.1±0.1 ^a	20.9±0.2 ^a	14.4±0.1 ^a	16.1±0.2 ^a
					TOF	11.7±0.2 ^a	12.0±0.2 ^a	15.1±0.2 ^a	14.4±0.4 ^a
					TPC	90.1±0.2 ^a	93±1 ^a	85.5±0.4 ^a	85±1 ^a

TPA-Total phenolic acids, TP -Total procyanidin, TOF-Total other flavonoids, TPC-Total phenolic compounds



Graph 1. This graph represents the assay done in triplicate, where the results are the average of the inhibition percentages. The graph compares the inhibition percentages obtained in the extract samples with respect to the control.



Concluding Remarks

- The results noticed a positive relation between phenolic compounds content and antimicrobial capacity of the leaves raspberry's extracts.
- The findings reveal a total antioxidant activity of 93.5%±0.12.
- The extracts exhibited antibacterial activity against all *K. pneumoniae* strains from clinical isolates, with inhibition rates equal to or exceeding 50%.
- These findings suggest that aqueous extracts of raspberry leaves may serve as a valuable therapeutic resource for combating multi-resistant bacteria.



References

[1] Garcia, J., Rodrigues, F., Castro, F., Aires, A., Marques, G., & Saavedra, M. J. (2022). Antimicrobial, Antibiofilm, and Antioxidant Properties of Boletus edulis and Neoboletus luridiformis Against Multidrug-Resistant ESKAPE Pathogens [Original Research]. *Frontiers in Nutrition*, 8.



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