ANTIMICROBIAL ACTIVITY OF MEDICINAL MUSHROOM: ALGERIAN DESERT TRUFFLE

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Abstract - Tirmania pinoyi (Maire)Malençon is a famous edible fungus in Algeria, well-known for its pleasant flavor and aroma. It is consumed as food and for medicinal properties. The aim of present study is to investigate the antimicrobial activity of T. pinoyi collected from El Bayed in West of Algeria. Soxhlet apparatus was used for extraction with a series of solvents, dichloromethane, chloroform, ethyl acetate and methanol in sequence of increasing of polarity. The results obtained revealed that the nature of solvents had a significant effect on both the antioxidant and antimicrobial activities. The antimicrobial activity of T. pinoyi was evaluated against two Gram-positive and two Gram negative bacteria using agar well diffusion methods. The results showed that the majority of the extracts investigated showed greater activities against the Gram-positive and negative bacteria.

Keywords - Desert truffle, Tirmania pinoyi (Maire), Antioxidant activity, Antibacterial activity, DPPH, well diffusion methods

I. INTRODUCTION

T. pinoyi (Maire) Malençonbelongs to the family of Terfeziaceae, order Pezizales. It is an edible mycorrhizal fungus, their geographical distribution is limited to arid and semi-arid lands, especially in North-Africa and Middle East. Several studies on the chemical composition of desert truffles showed thatthe protein content, which averages 20% of the dry weight in desert truffles, is significantly higher than in most vegetables and other fungi and, therefore, the consumption of these truffles is recommended (Bukhary and Parvez, 1993; Murcia et al., 2003). Desert truffles comprise a vast unexploited source of therapeutic compounds with anti-inflammatory, immunesuppressor, antimutagenic and anticarcinogenic potential (Hannan, Al-Dakan, Aboul-Enein and Al-Othaimeen, 1989). Traditionnel uses of desert truffles have motivated our effort to investigate the antimicrobial activities. Therefore, the aim of this study is to evaluate the antibacterialof different extrats of T. pinoyi.

II. MATERIALS AND METHODS

2.1. Truffle materials

Fresh T. pinoyi (Maire) Malençon scocarps, with no apparent physical or microbial damage, were collected from El Bayed and were authenticated by Pr. FORTAS, University of Oran, Algeria. All the truffle samples were identically selected in terms of size, shape, colour, and ripening stage. The fresh truffles were cleaned from soil anddried in the shadow, they were then ground into fine powder and stored at ambient temperature in a dry place and in the dark until use.

2.2 Soxhlet extraction.

The dried truffle powder of T. pinoyi (100 g) was successively extracted by Soxhletapparatus using solvents of increasing polarity as follows: dichloromethane, followed by chloroform, ethyl acetate and finally with methanol during 24 h for each solvent. The volume of each solvent used is 500 mL which was then evaporated under vacuum using a rotary evaporator and then weighed to calculate the yield of the extracts and stored at 4°C until required.

2.3 Antimicrobial assay

2.3.1. Microbial strains

Microbial cultures of four different strains of both Gram positive and negative bacteria were used for the determination of antimicrobial activity. Two Gram-positive (Staphylococcus aureus ATCC 6538, Enterococcus feacalis ATCC 2035) and two Gram-negative (Escherichia coli ATCC 25922, Pseudomonas aeruginosa ATCC 14028).

2.4 Determination of antibacterial activity

The antimicrobial activity of T. claveryi extracts was determined using agar well diffusion method following published procedure with slight modification (Perez et al., 1990). Each bacterial suspension was spreaded over the surface of Muller-Hinton Agar plates. The plates containing wells of 6 mm diameter were filled with 30µL extracts. The plates were then incubated at 37°C for 24 h. The results were expressed in terms of the diameter of the inhibition zone. All tests were performed in triplicates.Streptomycin (30 µg) was used as positive control.

2.5 Statistical analysis

Excel (Microsoft Corporation, USA) and STATISTICA (version 10) were used for statistical analysis. Data are expressed as means ± SD. To assess the variation of the variables among samples, a two-way Analysis Of Variance (ANOVA) was performed. Statistical significance between means

was determined using Duncan's multiple range tests and set at $p < 0.05$.

III. RESULTS AND DISCUSSION

In this study, the antibacterial activity was tested with the dichloromethane, chloroform, ethyl acetate and methanol extracts of T. pinoyi. The results of the agar well diffusion method using different polarity solvents were shown in Table.1.

Table 2. Screening of antimicrobial activity ofT. pinoyiin comparison with standard antibiotic.

Microorganisms	Inhibition zones diameter (mm)				
	Dichloromethane	Chloroform	Ethylacetate	Methanol	Streptomycin30µg/disc
Gram-positive bacteria Staphylococcus aureus ATCC6538	18±1[a]	13±1[a]	14±1[a]	13±1[a]	40.66±0.57
Enterococcus faecalis ATCC2035	12.33±0.58[c]	13±1[a]	06±0[b]	06±0[c]	27.5±2.88
Gram-negative bacteria Pseudomonas aeruginosa ATCC14028	13±1[cd]	20±1[d]	15.33±0.57[a]	10.33±0.57[e]	38.75±1.5
Escherichia coli ATCC25922	15.33±0.58[cd]	06±0[e]	15±1[a]	06±0[c]	37±2.64

Values, including diameter of the filter paper disc (6.0 mm), are means of three replicates.Mean values within each column followed by different letter differ significantly at ($p>0.05$).Values within the same column followed by the same letter are not significantly different ($p<0.05$). Among the different extracts, the chloroform extract of T.pinoyi showed significantly high antimicrobial activity against both Gram-positive and Gram-negative bacteria than the other extracts ($p<0.05$). The largest zone of inhibition (20±1 mm) was found to be exhibited against P. aeruginosa.The dichloromethane, chloroform and ethyl acetate extracts showed appreciable antibacterial activity against most bacteria tested as stated by Hamza et al., (2013). However, the methanolic extract hada moderate activity. Furthermore,we observed that the nature of solvents used has significantly ($p<0.05$) an impact on the antimicrobial activity of T. pinoyi.

CONCLUSION

In the present work, it had been concluded that different solvents used had a significant impact on the antimicrobial activity of T.pinoyi. Moreover, it was noted that different extracts of T.pnoyishowed marked antimicrobial activity activity revealing their potentials for therapeutic uses. However, the identification, characterization and purification of their bioactives compounds will be the aim of further investigation.

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