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PHYTOCHEMICAL SCREENING AND ANTIFUNGAL ACTIVITY EVALUATION OF GUM ARABIC (*ACACIA TORTILIS* FORSSK)

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Abstract

Studies on the *Acacia tortilis* (*Forssk*) species are numerous due to the ecological, economic and social importance of this species in several countries. For this, the aim of our work is the evaluation of the antifungal effect of the aqueous extract of gum Arabic exuded from *Acacia tortilis* (*Forssk*) and an attempt to characterize these main constituents by biochemical methods (screening phytochemical by reactions color and by thin layer chromatography).

The results of this study reveal a wealth of gum in flavonoids, tannins, sterols, oses…

The antifungal power of the aqueous extract based on gum Arabic has shown that the appearance of the zones of inhibition is proportionally linked to the increase in the dilution of the extract (1/100, 1/250, 1 / 500, 1/1000, 1/5000). However, *Fusarium oxysporum* have a resistance compared to other strains of mold.

Key words: Arabic gum, *Acacia tortilis*, antifungal activity, phytochemical screening and TLC.

Introduction

Medicines always need the nature and its natural products for all their requirements (Jyoti Rani *et al.*, 2020). Medicinal herbs and their therapeutic benefits; not just folkloric beliefs we inherited for grandmothers and elders, it contains these herbs contain potent compounds that give them; it applied properties.

Accacia tortilis (*Talha*) is a tree with a height between 2 and 10 meters, with flowers of white, yellowish color and spiral horns. This tree is found flat in the entire Algerian desert, especially the west southern one.

The gum Arabic (Alk) is one of the medical products that you can get from the Talha (Fig. 1), Among the advantages of the gum Arabic is that it is a natural organic product that has no taste, color, smell, or calories and is characterized by not interacting with chemicals easily while it dissolves in water (Shaymaa Saady Lafta *et al.*, 2019; Shaymaa Saady Lafta *et al.*, 2019), so it is used in many industries such as textiles, medicinal drugs, canned foods, soft drinks and adhesives, Industrial oils, cosmetics, paints and many other industries are diverse...this is thanks to its chemical composition (Nasir *et al.*, 2012).

The chemical composition of the gum Arabic may vary slightly depending on its origin, climate, harvest season, tree age and processing conditions, such as spray dying (Al- Assaf *et al.*, 2005(a,b); Flindt *et al.*, 2005; Hassan *et al.*, 2005; Siddig *et al.*, 2005). To the best of our knowledge, there is no information and no studies of antimicrobial activities were performed for the gum of the Talha of the west southern of Algeria but a local ethnobotanical survey and composition carried out showed its possible antifungal activity.

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Material and Methods

Location and plant material

We have chosen to collect samples from the Tindouf area; because it is characterized by the abundance of our plant, the south western part of Algeria, at coordinates (28°16'03.6"N 8°06'24.4"W) and around it. The plant was identified by Pr. MOUSAOUI A., director of laboratory

Fig. 1: *Accacia tortilis* and (a) tree branch; (b) granules of gum Arabic (Source, 2020).

of plants' resources and food security of semi-arid areas of southern- west of Algeria University of Bechar, Algeria.

This choice has been based on its frequent use by the local population in traditional medicine.

Collection and preparation of samples

Gum Arabic is natural exudates collected from the trunk and branches of shrubs in two ways (natural or artificial): This secretion can occur naturally when the tree is injured (wind, animals, insects...) or artificially when Nan causes bleeding for the purpose of harvesting.

The trunks are notched to harvest the exudates for six months (from December to June).

Gum Arabic was collected, taken to the laboratory, cleaned with distilled water and wiped with a clean cotton cloth, then dried. Dried gum Arabic was crushed successively in a porcelain mortar and then in a hammer mill Culati brand. The powders obtained were saved using a 160µm sieve of mesh diameter and then chemically characterized.

Screening phytochemical

Phytochemical screening by means of color reactions and by thin layer chromatography (TCL) was produced according to the analytical techniques (Table 1) described in the work by (Godin, 1954; Ladiguina *et al*., 1983; Georgievskii, 1990; Dawson *et al*., 1991; Wagner and Bladt, 1996; Dekker, 2002; Chaaib, 2004; Lagnika, 2005; Mamyrbekova-Bekro *et al*., 2008; Divya Sharma *et al*., 2019).

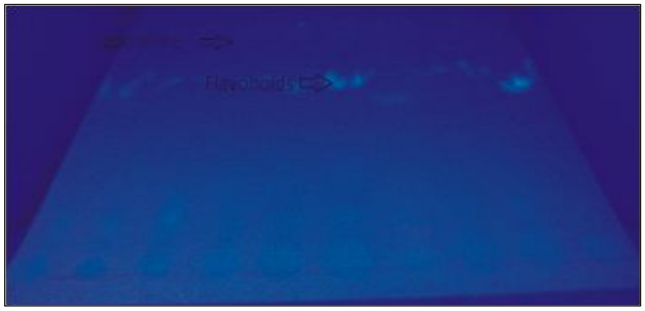

Fig. 2: Chromatograms for detecting flavonoids revealed by $AlCl_3$ in the visible (A) and under.

Extract preparation

The gum powder is dissolved in sterile distilled water using a vortex shaker to obtain the following concentrations (1/100, 1/250, 1/500, 1/1000, 1/5000).

Antifungal activity tests

• Fungal species:

Eight phytopathogenic fungi, including *Aspergillus flavus, Aspergillus parasiticus, Aspergillus ochraceus, Aspergillus fumigatus, Aspergillus niger, Penicillium expansum, Fusarium oxysporum* and *Alternaria sp* were these fungi cause different infections in many plants cultivates and stored products. The fungi were obtained from the collections of biology Department, Faculty of Science, Bechar University; Algeria.

• Culture media

Potato dextrose agar (PDA) was used (200g potato juice, 20g glucose, 15g agar-agar and 1000ml distilled water) amended with 40ppm chlorotetracycline hydrochloride and it used two for the determination of antifungal effect (Nuh Boyraz and Musa O¨ zcan, 2005).

• Determination of antifungal effects:

Antimicrobial tests are carried out according to the method reported by Remmal *et al*., (1993), Farah *et al*., (2001) and Satrani *et al*., (2007). The extract is emulsified with a 0.2% Agar solution in order to disperse the compounds and improve their contact with the germs tested.

Dilutions are prepared 1/10, 1/25, 1/50, 1/100, 1/500 in this Agar solution. In test tubes, each containing 13.5 ml of PDAac medium, 1.5 ml of each of the dilutions was added to obtain the final concentrations of 1/100, 1/ 250, 1/500, 1/1000, 1 / 5000, then shake the tubes before

Table 1: Characterization reaction of phytochemical screening.

Chemical components	Alkaloids	Flavonoides	Saponosides	Tanins	Reducing compounds	Terpens
Coloured reactions	Reagents of dragendroff and mayer	Reaction of cyanidine	Foam index	Ferric chlorides	Reagent of fehling sterols	Reagents of liebermann buchard

Table 2: Phytochemical screening of gum Arabic by color reactions.

Chemical groups	Flavo-noids	Quinon	Tanin	Sapon-oside	Alkaloid	Steroid and Terpene	Coumarin
Results	++	+	+	+++	+/-	+	+/-
+: Presence; -: not detected							

pouring them into Petri dishes. Controls containing the culture medium and the 0.2% Agar solution alone are also prepared. Seeding is done by touching using a platinum handle. The reading is done after four to seven days for mushrooms. Each test is repeated three times (Bourkhiss *et al.*, 2007).

• Evaluation of antifungal effects:

In order to evaluate the inhibition of mycelial growth of fungi by hydro-sols, the fungi colony diameters were measured since the day of incubation (Leroux and Gredet, 1978). The inhibition percentages of hydro-sols were calculated according to the following formula (Deans *et al.*, 1990):

$$I = \frac{(C - P)}{C} x100$$

I: inhibition (%); C: diameters of colonies on control Petri dishes; P: diameters of colonies on practiced Petri dishes.

Results and Discussions

The results of the phytochemical screening by color reactions

The phytochemical screening of gum Arabic makes it possible to demonstrate the presence of certain chemical groups; flavonoids, saponins, tannins, sterols and terpenoids (Table 2).

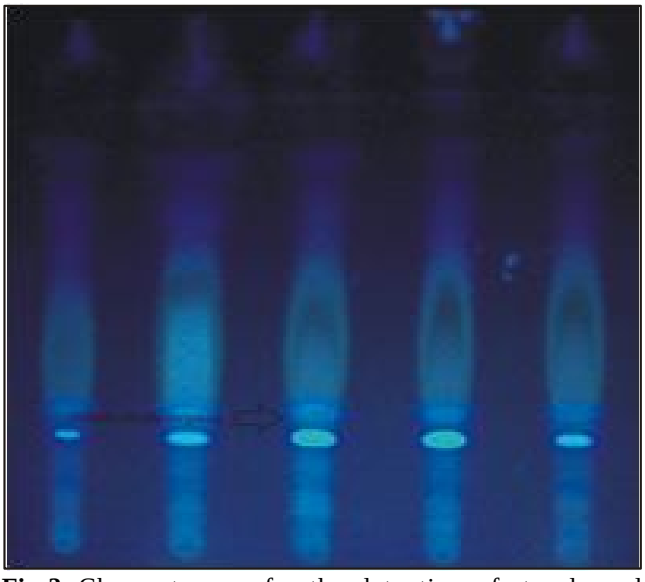

Fig. 3: Chromatogram for the detection of sterols and polyterpenes revealed by the Liebermann reagent.

The data obtained during phytochemical screening by a methodology are based on the detection of compounds by means of protocols oriented according to the results obtained during biological screening. They are often consistent with those of the literature (Adomi and Umukoro, 2010; Mujovo, 2008; Krasniewski *et al.*, 2006; Djouossi *et al.*, 2015; Mahalakshmi *et al.*, 2020).

Phytochemical screening by TLC

The results of the phytochemical screening by TLC of gum Arabic are presented in figures (two, three and four). The orange, yellow, blue, green, pink, purple spots observed on the chromatogram under UV/366nm may correspond to several classes of secondary metabolites. In order to specify the nature of the compounds revealed at UV/366nm, the specific reagents for coumarins (KOH, (CH_3CO_2) 2Pb, NH3); flavonoids (Neu reagent, $AlCl_3$, NH_3); sterols and polyterpenes (Libermann-Bürchard and Godin reagents); alkaloids (Dragendor'ff reagent) and tannins ($FeCl_3$) were used.

The results of this screening make it possible to develop hypotheses to explain the biological activity of an extract by the presence of a particular chemical family.

Alkaloids according to Hadjimi, (2011); affect the appearance of the fungal colony and cause a decrease and / or inhibition of mycelial which growth is variable depending on the concentrations tested.

Terpenes and their derivatives are promising in general used in antifungal therapies (Cox *et al.*, 2000; Inoue *et al.*, 2004).

In vivo, a good number of saponosides defend the plant against microbial or fungal attack. Bruneton, (1999) and Kurkin, (2003) have shown that simple phenol, phenolic acids and flavonoids have anti-inflammatory and anti-hemorrhagic properties, antibacterial and antifungal properties, in particular with regard to phytopathogenic organisms.

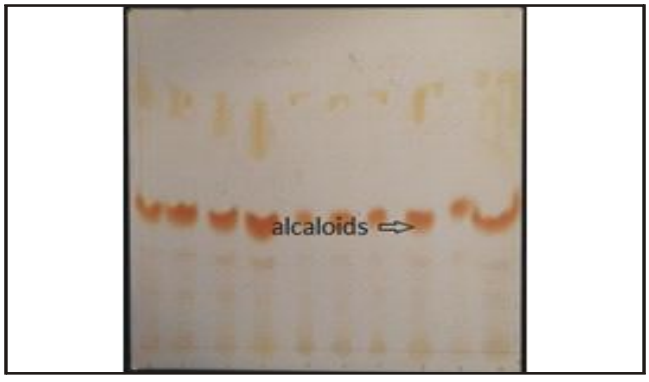

Fig. 4: Alkaloid detection chromatogram. Developer: $CHCl_3$/$(CH_3)_2CO$/$(C_2H_5)2NH$/Ether.

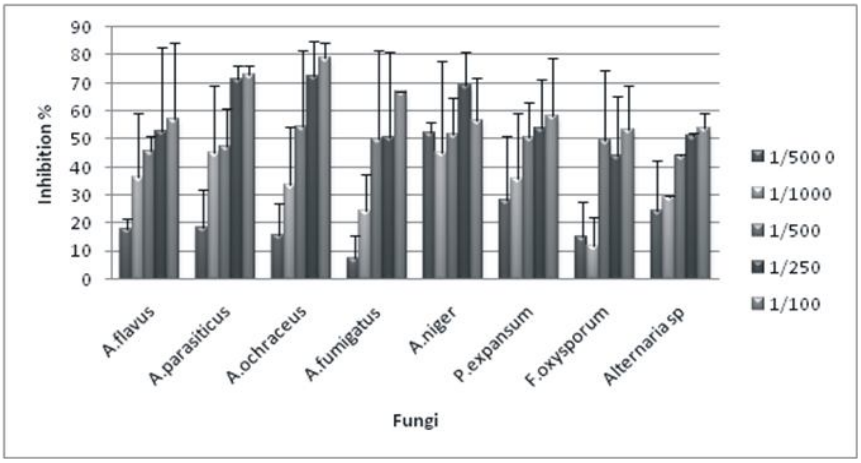

Fig. 5: The antifungal activity of gum Arabic.

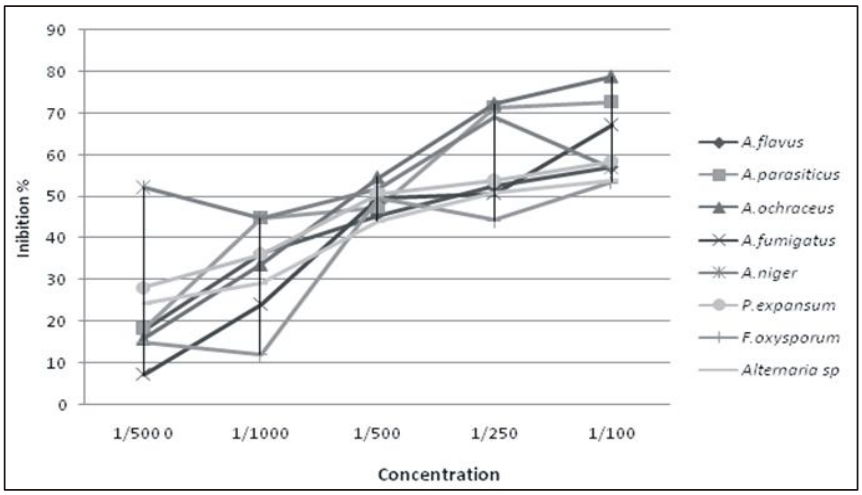

Fig. 6: Correlation of the antifungal activity of gum Arabic.

Many quinones are antibacterial and fungicidal (Riffel *et al.,* 2002).

Antifungal effect

The antifungal activity of gum Arabic extracts was evaluated by the method of diffusion in agar medium. All dilutions have antifungal activities on all the strains of mold tested (*Aspergillus flavus, Aspergillus parasiticus, Aspergillus ochraceus, Aspergillus fumigatus, Aspergillus niger, Penicillium expansum, Fusarium oxysporum* and *Alternaria* sp).

Inhibition values exceed 50% for all mold strains from dilution (1/250) except *Fusarium oxysporum*. It proves to be the most resistant for all the concentrations tested, this is linked to its great ability to develop resistance to many antimicrobial agents, where several authors actually report the low sensitivity of this strain against plant extracts (Baayen and Elgersma, 1985). We noticed a convergence of inhibition values at the 1/500 concentration in all the strains tested.

The antifungal activity of the aqueous extract of gum Arabic is probably effective thanks to the various chemical agents present in this polysaccharide exudate y including saponin, essential oils, tannin, flavonoids, polyphenols and alkaloids and the high terpene content (Chaubal and Tambe, 2006; Wisdom and Shittu, 2010). Gum Arabic also contains several types of enzymes such as oxidases, peroxidases and pectinases, some of which have antimicrobial properties (Fig. 5, 6) (Saini *et al.*, 2008).

Conclusion

Gum Arabic is used in food industries, cosmetic science, clinical and pharmacy as an oxidant, anti-microbial, anti-coagulant, anti-inflammatory and shelf life enhancer of food products.

Through the results obtained, it has been demonstrated that gum Arabic has a more or less interesting antifungal potential. Consequently, it could be a source of metabolites with drug and pharmaceutical interests.

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