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Evaluation of antifungal effect and non-growth diameter of Persian oak's Jaft extract on *Penicillium Digitatum* and *Penicillium Italicum* molds (indicator of mold spoilage after citrus fruits harvest

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ABSTRACT

This study aimed to evaluate the antifungal effect of Iranian oak Jaft extract on *Penicillium italicum* and *Penicillium digitatum*, as an indicator of mold spoilage after citrus fruits harvest. Different extraction methods were used to obtain tannins and flavonoids from the Oak Jaft. The study investigated the Minimum Inhibitory Concentration (MIC), Minimum Fungicidal Concentration (MFC), and non-growth diameter of the molds using three different methods: tube dilution, surface culture, and diffusion in agar well. The results showed that different concentrations of aqueous extract had varying effects on the non-growth diameter of the molds. The antifungal effect was significantly increased with a concentration of 40 and 80 mg/ml of aqueous extract. In comparison, the control group, which used Fludioxonil 25% poison, showed a significant difference in mold growth. Therefore, the extract of Iranian oak Jaft can be used as an alternative to control fungal spoilage of citrus fruits.

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1. Introduction

In recent years, public awareness of the side effects of using chemical fungicides, improvement of safety and quality standards, etcAlso, the emergence of strains resistant to chemical drugs and the proliferation of resistant microbial isolates due to the excessive use of antibiotics.The desire to consume natural products has increased and efforts are being made By discovering natural antimicrobial agents that are compatible with the environment and living organisms,Like essential oils and plant extracts, a suitable replacement for antibiotics and chemical poisons can be obtained. [1]In this regard, extensive research is being done to increase the scope of new indigenous herbal medicines, which have a special strategic, productivity and economic value in the world. Since the country of Iran has a lot of useful and valuable plant diversity in terms of biological ecosystem, identifying and investigating the physicochemical activity of indigenous plant materials (oils, essential oils, tannins, etc.), plays an important role in the optimal use of national wealth. Iranian oak extract has long been recommended in traditional Iranian medicine for the treatment of various diseases. The study of scientific sources shows that in recent years, various laboratory researches have been conducted in the field of investigating the effect of this plant extract on plant pests, fungi and bacteria, which discovered a similar inhibitory effect.Oak fruit extract on bacteria*Shigella flexneri*[1] compared to ciprofloxacin antibiotics[2] and trimethoprim sulfomethoxazole[3] [2], the effect of tannins in oak extract on the treatment of digestive damage and water absorption and protein deposition in the intestine [3], the lethality of hydroalcoholic extract of oak placentaSaprolegnia mushroom[4][4] and preventing its growth compared to the antifungal drug Malachite Green[5] [5], the inhibitory effect of the alcoholic extract of the placenta of the oak plant on Candida yeast due to production A secondary metabolite called lavanoides[6] at that [6]، Comparison of the antibacterial activities of thyme, oak sap and hydroalcoholic extract of pistachio green shell on L. monocytogenes[7] And the highest antibacterial effect of placenta on this bacteria [7]، Inhibitory effect of alcohol extract of placenta on Acinotobacter[8] [8], similar effect of oak fruit hydroalcoholic extract with gentamicin[9], lower effect than kanamycin[10] and higher effect than tobramycin[11] on Staphylococcus aureus[12] And also its lower effect compared to gentamicin and kanamycin on Escherichia coli, but more effect than tobramycin on A.n [6]Production of herbal mouthwashes with antimicrobial activity containing extracts of oak bark and Shiraz thyme[9], Aare among the achievements of this research. Oak is one of the plants of the mountainous regions of the world, with a great variety of species. Iranian oak with the scientific name of Queros Branty[13] and the Latin nameThirty one[14], from the Fagasse family[15], is one of

[1]- Shigella Flexneri

[2] -Ciprofloxacin

[3]- Trimethoprim

[4]- Saprolegnia

[5]- Malachite green

[6]- Flavonoids

[7]- listeria monocytogenes

[8]- Acinetobacter

[9]- Gentamicin

[10]- kanamycin

[11]- Tobramycin

[12]- Staphylococcus Oreos

[13]- Brantius oak

[14]- Brantii Oak

[15]- Fagaceae

the dominant species in the north, southeast of the Zagros mountain range and the central regions of Iran [10]. According to the studies, the height of this tree plant is approximately 20 meters. Its leaves are usually uniform and egg-shaped with a threaded margin and grow from the base. These leaves are shiny and dark green in color and their surface is a little rough. Oak fruit called acorn[16] In a bowl called Gland[17] contract. These fruits have different amounts of oily substances, different sugars, starch, and a small amount of quercetin[18], pentosan and tannin. The shape of Iranian oak fruit is smooth and oval and consists of two parts, cup and capsule, and the capsule also has three parts. The outer shell that surrounds the capsule and accounts for about 7% of the weight of the fruit (about 15 to 20 grams) [11]. The placenta is a thin shell that is wrapped around the brain. Oak skin and fruit are rich in polyphenolic compounds, and in addition to its antimicrobial properties, it is used in the treatment of chronic skin diseases, eczema, varicose veins, bleeding, stomach ulcers, hemorrhoids, tonsillitis, edema, and poisoning caused by alkaloids.[12]. Glycosides, alkaloids, tannins, saponin, flavonoids, resins, terpenes, steroids, phenolic acids, protein, fiber, minerals and vitamins A, B and C are the compounds found in different parts of oak fruit.[13].

Genus Penicillium from Hyphomycetes[19](a group of incomplete fungi). Penicilliums are the largest group of soil fungi, with more than 150 species. These fungi are one of the most important food spoilage factors. The optimum growth temperature of these mushrooms is between 22 and 27 degrees Celsius. Two important species of this genus, including Penicillium digitatum and Penicillium italicum, under the name of green and blue molds, respectively, play a major role in the spoilage and decay of citrus fruit after harvesting in storage. [14]. This laboratory research is an attempt to evaluate the activity of an extractable and available product obtained from natural and plant resources of the country, which can provide the basis for reducing the consumption of chemical fungicides during the storage of fresh citrus fruits. In this study, the antifungal effect of oak sap extract on strains of Penicillium digitatum and Penicillium italicum (green mold and blue mold of citrus fruits) was investigated by quantitative and qualitative methods.

2- materials and methods

2-1- Preparation of raw materials and preparation

In this research, a pair of Iranian oak was purchased from an attari in Khorram Abad city. After confirming the scientific name of the oak plant and cleaning and removing its dust by blowing, using an electric mill, mill, and its powder was prepared. In order to prepare the plant extract, several methods were used;

2-1-1-extraction by soaking method (maceration[20])

Polar and non-polar solvents were used for extraction. In the first method, first, 100 grams of oak sap powder was soaked with 500 ml of distilled water and 96% ethyl alcohol in an equal ratio in an Erlenmeyer flask and kept in the dark for three days. During the storage period, the contents of

[16] -Acorn

[17]- Gland

[18]- Quercetin

[19] - Hyphomycete

[20]- Maceration

the jar were stirred with a shaker every day for 20 minutes. After 72 hours, the contents of the Erlenmeyer flask were filtered with a smooth multilayer sterile gas and the extracted extract by a centrifuge (model3-16PK Sigma, Germany) was centrifuged at 3000 rpm for 10 minutes. The clear extract (upper phase) obtained from the centrifuge was transferred to the round-bottom Erlenmeyer of the rotary evaporator and extracted at a temperature of 60 degrees Celsius under vacuum conditions. In order to completely evaporate the water of the concentrated extract in the device, it was placed in the oven at a temperature of 40 degrees Celsius and dried and kept in the refrigerator until use. [4].

In the second method, diethyl ether along with distilled water was used to soak oak sapwood powder, this process was done to separate fats, vegetable pigments and resins. According to the method mentioned in the first method, the contents inside the Erlenmeyer flask were kept in the dark for 72 hours and after straining, the obtained liquid was dried in a rotary evaporator and oven and kept in the refrigerator until use. [15].

2-1-2- Extraction by boiling method

In this method, placenta powder was boiled in a specific volume of water. In this method, which is suitable for extracting effective substances that are soluble in water and thermally stable, the ratio of plant to water was fixed (1:4) and the process of boiling water and extracting the extract continued until the volume of the solvent reached a quarter of the initial volume. Found. After cooling, the concentrated extract was smoothed with multi-layered sterile gas and transferred to the round-bottomed Erlenmeyer of the rotary evaporator and extracted at a temperature of 60 degrees Celsius under vacuum conditions. The concentrated extract in the extractor was dried in an oven at 40°C for complete evaporation of water and kept in the refrigerator until use.

2-2- Preparation of mushroom strains

1-2-2- Green mold

strains (ATTC 481130) and blue mold (ATTC 488140) was prepared and used from the citrus research center of Gilan province, Rasht city. These molds are non-pathogenic and can be cultivated on nutrient agar culture media. To prepare the fungal suspension, by the method mentioned in the Iranian National Standard No. 2325, the spores of the mushrooms were removed from the culture medium by a sterile culture ring and transferred to a tube containing a salt solution containing polysorbate. The resulting suspension was filtered using a sterile non-absorbent membrane filter and the number of spores was counted using a blood cell count slide (Neobar). The number of spores is $10^{\vee}$3.5 x spores per milliliter were adjusted and stored in microtubes in a freezer at a temperature of minus 70 degrees Celsius. [16].

2-2-2- Determining the number of microorganisms that can grow in the suspension

Using Ringer's lactate solution, successive tenfold dilutions were prepared from the prepared suspensions of both molds. Then, from the dilutions that were expected to have a countable number of colonies on the plate after cultivation, they were cultured on subrodextrose agar culture medium by the surface culture method and the plates were placed in a greenhouse at a temperature of 25 degrees Celsius for 3 to 5 days. The number of colony-forming units was determined using the following formula[16].

$$N' = \frac{\sum C}{V \times n \times d}$$

∑C total number of colonies counted on two plates, one of which contains at least 10 colonies;

V volume of inoculum used in each plate in millimeters;

n is the number of selected plates, in this case n

2-3- Evaluation of the antifungal activity of the herbal extract of Jaft Balut in laboratory conditions

To determine the antifungal activity of the plant extract of oak placenta by the tube dilution method to determine the minimum inhibitory concentration.MIC(and the surface culture method to determine the minimum lethal concentration of the fungus)MFC(a) The methods mentioned in the Iranian National Standard No. 5875 and the well release method)WD) used[17].

2-3-1-Determining the minimum growth inhibitory concentration(MIC)

At first, dilutions of plant extracts were prepared. To prepare the first dilution, 8 mg of the extracted extract was mixed with 10 ml of distilled water and mixed with a stirrer. Due to the fact that during the test, 1 milliliter of the desired fungal suspension was added to each tube, and as a result, the prepared dilution was reduced to half, so from the beginning, each tube corresponding to each dilution was considered to contain twice the desired concentration.،became. To prepare successive dilutions of 1:2, one milliliter of the first dilution prepared from the plant extract was added to the next tube containing 1 milliliter of Sabro dextrose broth culture medium. One milliliter of this tube was added to the next tube containing 1 milliliter of culture medium and continued in the same order until it reached the lowest acceptable dilution. Then 1 milliliter of the last tube was removed and discarded. In this way, each tube contained 1 milliliter of plant extract diluted in culture medium[17]. To determine the minimum inhibitory concentration of Penicillium digitatum and Penicillium italicum mold strains, 8 sterile test tubes were placed separately for each spore. 6 test tubes containing different concentrations of the extract respectively 8٪, 4%, 3%, 2%, 1.5%, 1%, 0.75% and 0.5%. Two test tubes were considered as positive control and negative control. of the initial suspension with concn CFU/ml $۱۰^{۷}$ 3.5× suspension of both blue and green mold spores with concentrationCFU/ml $۱۰^{۴}$3.5 × was prepared. 1 ml to the tubesliter of Subra dextrose broth medium and 1 mlA liter of mushroom suspension was added and after adding 1 ml of the dilutions of the prepared extracts, it was placed in a 25°C incubator for 24 hours. The minimum inhibitory concentration was determined.

۲-۳-۲- Determination of the minimum lethal concentration of the fungus (MFC)

To determine the minimum lethal concentration of oak placenta extract, test tubes in which turbidity was not observed (minimum inhibitory concentration tube and higher concentrations) were cultured on plates containing Subra dextrose agar culture medium and after being placed in a greenhouse at a temperature of 25 degrees Celsius after a period of 72 Hour, plates containing microorganism are examinedtook The first plate contains the lowest concentration of the extractin which the mold did not grow was considered as lethal concentrationbecame. The tests were repeated three times[17] (Table No. 1).

Table 1 Minimal Inhibition Concentration(MIC) of oak's jaft extract on molds

	Aqueous extract	Hydroethric and Hydroalcoholic extract
Mold	MIC (mg/ml)	MIC(mg/ml)
Fingered pencil	15	-*
Italian pencil	10	-

(-) Antifungal activity

2-3-3-Determining the diameter of the lack of growth halo

In the diffusion method in the well to determine the diameter of the halo of non-growth, standard laboratory and clinical guidelines (CLSI[21]) used. In this method, in each 9 cm petri dish, a mixture of one milliliter of fungal suspension, with an equivalent concentrationCFU/ml ۱۰۴x 3.5 with twenty milliliters of Sabro dextrose agar culture medium by surface culture method[22]For each of the strains, it was spread and by digging a well around and on the culture medium with a sterile pipette (pipette number 5 and depth of 4 mm), 30 microliters of the extracts in the concentration that was determined in the method of determining the minimum inhibitory concentrationMIC) had inhibited the growth of the desired fungi, it was poured into the wells by the sampler and kept at a temperature of 25 degrees Celsius for 72 hours. To compare the growth inhibitory power of sterile distilled water (plant extract solvent) as a negative control without extract and 0.25 ppm of fludioxonil 25٪ was used as a positive control. After three days, the culture media were examined under the laboratory hood and the diameter of the microorganism non-growth halo was measured using a ruler and the results were reported in millimeters. All tests were repeated three times (Tables 2 and 3).

[21]- Clinical and Laboratory Standards Institute

[22]- For Plate

Table 2 Results of MIC, MFC (mg/ml) measurement and average of non-growth diameter (mm) of Aqueous oak's jaft extract on *Italian brush*

Mold	Dilution of the Extract mg/ml	Extract concentration%	Average of non-growth diameter/mm	MIC* mg/ml	MFC mg/ml
Italian brush	80	8	24	-*	-
	40	4	22	-	-
	30	3	18.9	-	-
	20	2	18.5	-	-
	15	1.5	11.5	-	-
	10	1	19.5	-	+
	7.5	0.75	15.6	+	+
	_5	0.5	14	+	+

* Minimal Inhibition Concentration, *Minimal fungicidal Concentration., +,grow; -, not grow; n = 3.

Table 3 Results of MIC , MFC (mg/ml) measurements and average of nongrowth diameter(mm) for aqueous oak Jaft extract on *Digitized pencil*

Mold	Dilution of the Extract mg/ml	Extract concentration%	Average of nongrowth diameter/mm	MIC* mg/ml	MFC* mg/ml
Digitized pencil	80	8	21	+*	-
	40	4	21	+	-
	30	3	18	+	-
	20	2	18.33	+	-
	15	1.5	17	+	-
	10	1	13.77	-	-
	7.5	0.75	13.33	-	-
	_5	0.5	13.67	-	-

* Minimal Inhibition Concentration, *Minimal fungicidal Concentration; +, grow; -, not grow; n = 3.

2-3-4-Statistical analysis

Data by softwareSPSS 25 versions were analyzed.The effect of aqueous extract concentration on the diameter of the non-growth halo of both molds using the tableANOVA review andMean data obtained with Duncan's multiple range test at the probability level ۰٫۰۵ $p<$ To The title of significant differencecompared.

3-Results and discussion

1-3-minimum concentrationGrowth inhibitory concentration (MIC) and minimum lethal concentration of fungi (MFC)

Table 1. Minimum concentration resultsgrowth inhibition (MIC) Extracts of oak pairs extracted on blue and green penicillium molds at the maximum concentration tested (8٪) is showing. The results indicate that the hydroalcoholic and hydroether extract They do not have an antifungal effect on both molds, but the aqueous extract of oak sap is effective on the growth of both tested molds. The lowest concentration of aqueous extract inMIC equivalent to 10 mg/ml, on the mold Penicillium italicum with the diameter of the non-growth halo of 24 mm and on the mold Penicillium digitatum inMIC The equivalent of 15 mg/ml was obtained with the

maximum diameter of the non-growth halo of 21 mm (Tables 2 and 3).

2-3-Diameter of lack of growth halo

Measuring the diameter of the halo of non-growth is another method of determining the sensitivity and activity of microorganisms (molds) when faced with plant extracts. As can be seen in Tables 2 and 3, the lowest concentration of aqueous extract (mg/ml 5), on the mold Penicillium italicum with a halo diameter of 14 mm and the highest concentration of aqueous extract.mg/ml 80), the diameter of the aura of non-growth is 24 mm. Also, by increasing the concentration of aqueous extract, the diameter of the non-growth halo of Penicillium digitatum mold has also increased from 13.67 mm to 21 mm, and this amount has decreased compared to the diameter of the non-growth halo of Italicum mold.The results showed that the antifungal effect of Iranian oak placenta extract is concentration-dependent and has a significant effect on the diameter of the non-growth halo of green and blue Penicillium molds. Also compareAverage data obtained from Duncan's multi-range testThe probability level ($p<0.05$) indicated that changes in concentration have an effect on the diameter of the non-growth of molds and their difference is significant. There is the biggest difference in the concentrations of 40 and 80 mg/ml of the extract with other concentrations of the extract. In comparison, there was a significant difference ($p<0.05$) in the diameter of the mold growth halo in the control group that used 25% fludioxonil poison (Tables 4 and 5).

Table 4 Mean inhibition zone diameter (mm) of oak Jaft aqueous extract on Penicillium digitatum

Mean± SD	extraction groups mg/ml
45.0000[a]	Control +
22.8333±5.7 [b]	80
21.4583±6.2 [bc]	40
18.4583±3.6 [bcd]	30
16.0833±3.6 [cd]	15
15.4167±4.0 [d]	20
14.5000±1.1 [d]	7.5
14.0833±3.4 [d]	5
12.9583[d]±1.9	10

Values are expressed as mean ±standard deviations, n = 3; different letters (a, b, c and d) in each column show significant difference at $p < 0.05$

Table 5 Mean inhibition zone diameter (mm) of oak's Jaft aqueous extract on Penicillium italicum

Mean± SD	extraction groups mg/ml
45.0000[a]	Control +
22.8333±1.7 [b]	80
21.4583±4.3 [bc]	40
18.4583±2.1 [bcd]	30
16.0833±4.0 [cd]	15
15.4167±5.0 [d]	20
14.5000±3.7 [d]	7.5
14.0833±3.6 [d]	5
12.9583±2.2 [d]	10

Values are expressed as mean ±standard deviations, n = 3; different letters (a, b, c and d) in each column show significant difference at $p< 0.05$.

3-3- The relationship between the growth inhibition halo diameter and the concentration of the extract

Investigating the mutual effect of different concentrations of oak mate extract and two strains of Penicillium mold, green and blue, on the diameter of the halo of mold growth,

using analysis of varianceANOVA And Duncan's statistical test method showed that the concentration of the extract and the type of mold had a significant interaction effect on the diameter of the non-growth halo٠٫٥/ ٠ > *P* They do not have (Figures 1 and 2).

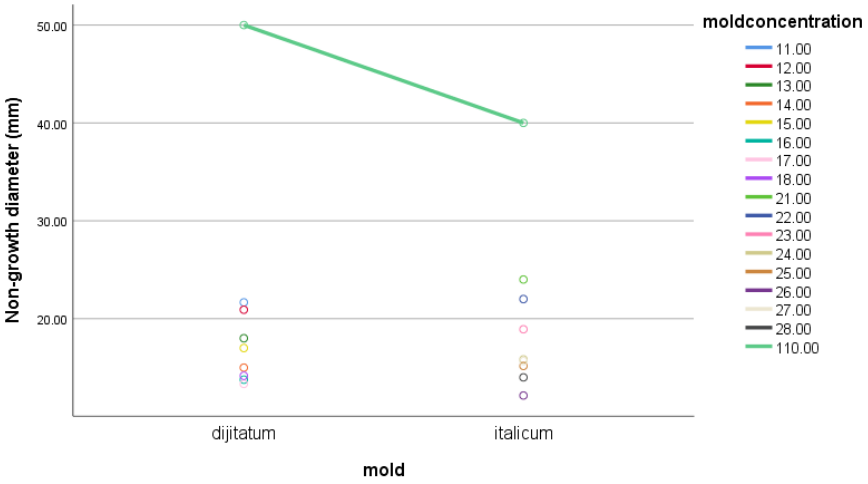

Fig 1 Concentration (mg/mm) versus molds & Estimated Marginal means of non-growth diameter in *Penicillium digitatum and Penicillium italicum*.

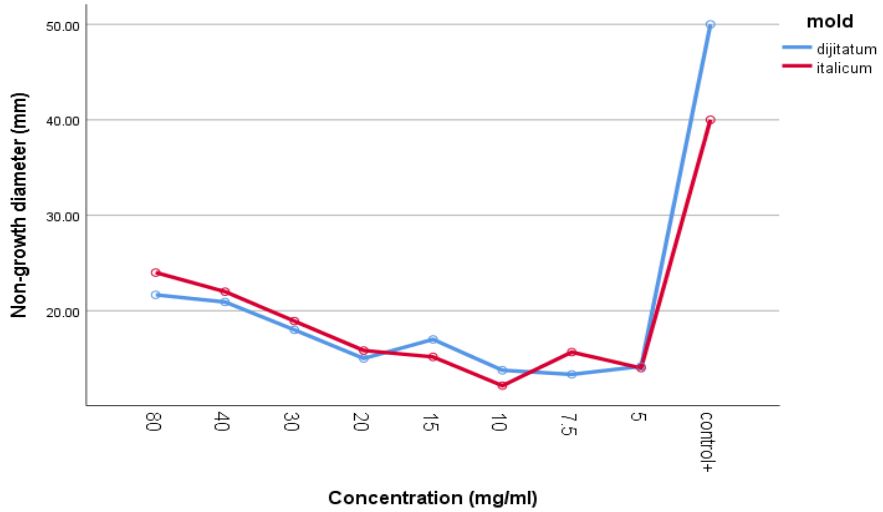

Fig 2 Estimated Marginal means of non-growth diameter with different concentrations of oak's jaft extract on *Fingered penci*land *Italian pencil*.

The mechanism of effect of the plant extract of the oak placenta can be attributed to the inherent antioxidant property of the constituents of plant extracts, which play an important role in the chemical defense system of plants against diseases. The most important phenolic compounds include phenolic acids, tannins, flavonoids [18]. Based on the plan of extracting various phenolic compounds from the skin of Kalberg and Kurth plants, it can be said that the substances in the hydroalcoholic and hydroether extracts extracted in this research include flavonoids and phlobatannins [19]. Phenolic compounds can be divided into simple phenols and polyphenolic compounds. Phenolic compounds are secondary metabolites known to have an aromatic ring containing a free hydroxyl

group. These molecules are present in almost all parts of plants and are involved in many physiological processes such as cell growth, root formation, seed germination and fruit ripening. Also, the antioxidant property of these compounds allows them to lose hydrogen and trap free radicals. It has been proven in scientific findings that phenolic metabolites in plants have the ability to cause free radical oxidation of fats and other biomolecules in the cell membrane of microorganisms and destroy them [20].For example, tannins, which are part of the components of oak placenta extract, can be toxic to bacteria, yeasts, filamentous fungi, and even viruses. By precipitating microbial proteins, tannins prevent their growth and can take food and proteins out of the reach of microbes or play a role through the mechanism of iron trapping, hydrogen bonding and specific distribution with vital proteins such as enzymes [21]. Tannins are able to inhibit the reverse transcriptase enzyme in human viruses to prevent their enzyme from multiplying [4].. This result with the results of Panahi et al[7] and Sharifi et al., who studied the effect of hydroalcoholic extract of oak sap on Candida albicus yeast and Saprolegnia mold [4] are not consistent, but in terms of proving the antifungal properties of its aqueous extract, the results of all the studies conducted so far on the antimicrobial effect of oak sap extract confirms (Figures 1 and 2). Based on the planKalberg and Korth, in the extracted water extract, some compounds soluble in polar and non-polar solvents (alcohol and ether) are not present, and despite their absence, the extracted water extract, unlike the aqueous extract and ether, is capable of inhibiting the growth and activity of molds. Penicillium and Digitatum. Therefore, it can be concluded that in the extraction process with alcohol and ether, some of the active ingredients of the extract, such as phenolic compounds and tannins, have reacted with alcohol and ether in the environment and have been deactivated. While in the aqueous extract, these compounds remain intact and play their biological role.

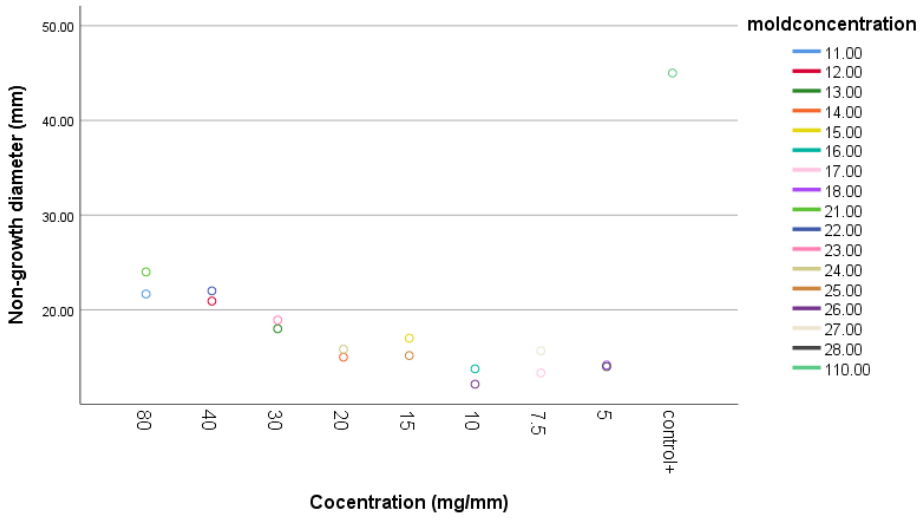

Fig3 Concentration (mg/mm) versus mold & Estimated Marginal means of non-growth diameter in *Italian pencil*

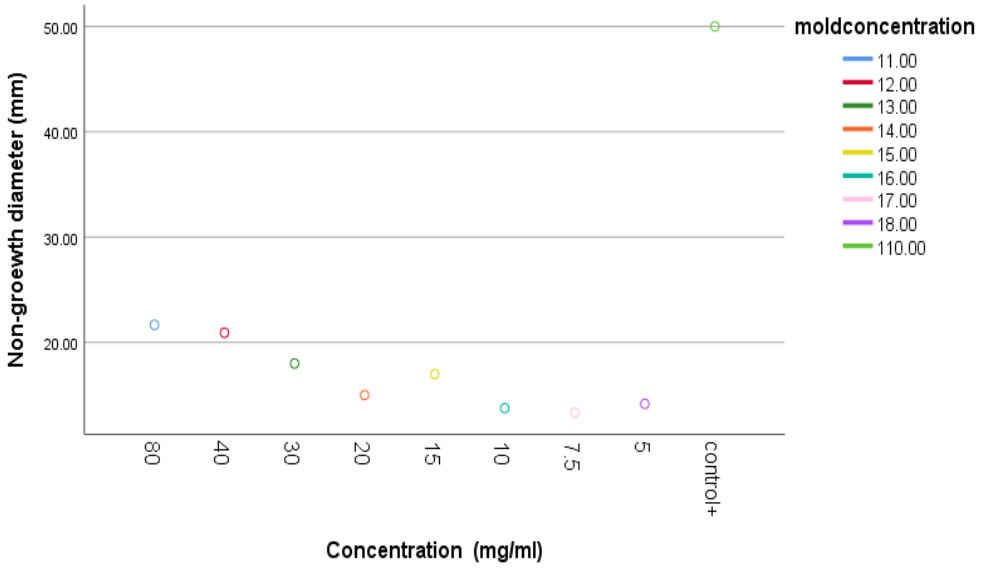

Fig 4 Concentration (mg/mm) versus mold & Estimated Marginal means of non-growth diameter in *fingered pencil*

4- Conclusion

The results of this research showed that the aqueous extract of oak sap has an antifungal effect on Penicillium digitatum and Penicillium italicum molds. The trend of human knowledge towards the use of herbal medicines is a way for the economic growth of most countries by producing native medicinal plants and extracting their effective substances in order to use organic substances in the industry. The production and extraction of these compounds with lower costs and their optimization makes it easier to achieve the desired goals. The use of aqueous extract of oak as an effective and cheap agent against molds that are an indicator of citrus spoilage can represent a bright horizon for reducing the waste of agricultural products and increasing their storage time.

It is suggested that in the future researches, the aqueous extract of the oak placenta should be investigated by chromatography methods and the main effective substances with the property of inhibiting the growth of the fungus or its lethality should be investigated and determined precisely.

5- Resources

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مقاله علمی_پژوهشی

ارزیابی فعالیت ضدقارچی و قطرهاله عدم رشد عصاره جَفت بلوط ایرانی برکپک‌های پنی‌سیلیوم دیجیتاتوم و پنی‌سیلیوم ایتالیکوم (شاخص فساد کپک‌زدگی پس از برداشت میوه مرکبات)

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چکیده

این مطالعه با هدف بررسی اثر ضدقارچی عصاره جفت بلوط ایرانی بر روی دو گونه کپک آبی (پنی‌سیلیوم ایتالیکوم۲۳) و کپک سبز (پنی‌سیلیوم دیجیتاتوم۲۴) به عنوان شاخص فساد کپکی میوه مرکبات انجام‌شد. برای به‌دست آوردن تانن‌ها و فلاونوئیدها از جفت بلوط از روش‌های مختلف استخراج استفاده شد. فاکتورهای مورد بررسی شامل حداقل غلظت مهارکنندگی[۲۵] (MIC میلی‌گرم بر میلی‌لیتر)، حداقل غلظت کشندگی[۲۶] (MFC، میلی‌گرم بر میلی‌لیتر) و قطر هاله عدم رشد۲۷(میلی‌متر) با روش‌های رقت لوله‌ای، کشت سطحی[۲۸] و انتشار در چاهک آگار (WD[۲۹]) تعیین شد. بررسی نتایج نشان داد، قطر هاله عدم رشد کپک‌ها در غلظت‌های مختلف عصاره آبی اختلاف معنی‌داری ($p<۰/۰۵$) دارند. اثر ضدقارچی در غلظت‌های ۴۰ و۸۰ میلی‌گرم بر میلی‌لیتر عصاره آبی به طور معنی‌داری($p<۰/۰۵$) افزایش‌یافت. در مقایسه، قطر هاله عدم رشد کپک‌ها در گروه شاهد که از سم فلودیوکسونیل[۳۰] ۲۵ درصد، استفاده شد، تفاوت معنی‌داری($p<۰/۰۵$) داشت. بنابراین می‌توان از عصاره جفت بلوط ایرانی به عنوان جایگزینی برای کنترل فساد قارچی مرکبات استفاده‌نمود.

[23] -Penicillium Italicum

[24]- Penicillium Dijitatum

[25] -Minimal Inhibition Concentration(MIC)

[26]- Minimal Fungicidal Concentration (MFC)

[27]- Non-growth Diameter

[28]- Surface Culture

[29]- Well Diffusion (WD)

[30] -Flodioxonil