

Morphocytological Effects of Two Home Spices on *Pisum sativum* Plants

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Abstract: Despite of *Thymus capitatus* and *Rosmarinus officinalis* are used for flavoring foods, food industry, antimicrobial, antifungal and antioxidant agents due to the presence of some important phenolic components in their essential oils. The present work showed the bad effect of these spices when used in large quantities which cause highly significant reductions in germination percentage, some morphological features, nucleic acids content and mitotic activity and also the appearance of high percentage of chromosomal abnormalities. This may be due to the poisonous effect of aromatic compounds that are found in these plants such as essential oils that include phenolic compounds and terpene-phenolic derivatives.

Key words: *Thymus capitatus* • *Rosmarinus officinalis* • *Pisum sativum* • Germination • Growth • Nucleic acids content • Mitotic activity • Abnormalities

INTRODUCTION

Spices tend to be aromatic or fragrant and have a pungent taste. Spices are generally produced from flowers, fruit, seeds, roots, leaves or bark. Thyme and Rosemary are the common spices in homes and used almost every day in food.

Thyme (*Thymus capitatus*) is a perennial plant commonly used as a spicy herb and locally known under the common name “zaa'tar” often used to flavor cough medicines [1]. Leaves are used in salads as garnishes and as flavoring for poultry, fish, beef, lamb, soups, herb butters, vinegars, beans and vegetables [2]. *Thymus* essential oils have been studied, mostly from the viewpoint of their flavor and fragrance chemistry, only for flavoring foods. Nowadays, however, essential oils and their components are gaining increasing interest because of their relatively safe status, their wide acceptance by consumers and their exploitation for potential multi-purpose functional use [3, 4].

Rosemary (*Rosmarinus officinalis* L.) has long been considered an important plant for its essential oil used in perfumes and medicine [5-7]. Rosemary belongs to family Lamiaceae and has been cultivated for a long time. Anthropologists and archaeologists have found evidence that rosemary herbs were used as medicinal, culinary and cosmetic virtues in the ancient Egypt, China and India. The essential oil enhances the blood-circulation of the limbs. It also has an antirheumatic effect and relieves the

neuralgic pains. Besides the therapeutical application, the essential oil is widely applied in the cosmetic industry producing various Cologne waters, bathing essences, hair lotions and shampoos. The leaf of rosemary is an indispensable spice of the French, Italian and Spanish cuisine [8].

MATERIALS AND METHODS

Seeds of *Pisum sativum* plant were kindly obtained from the Egyptian Agricultural Organization and Vegetable Department, Agricultural Research Center, Ministry of Agriculture, Giza, Egypt.

The seeds of *Pisum sativum* were individually pre-emergence treated with different concentrations of *Thymus capitatus* and *Rosmarinus officinalis*, respectively for 3 days and 6 days. The concentrations used were: 0%, 1% and 2%. Such range of the concentrations was chosen to be nearly equal that be used in cooking at home. The concentrations 1% and 2% were prepared by boiling 1 and 2 g of the plant material in 100 ml distilled water, respectively.

The seeds were planted equally in Petri-dishes (15 cm Ø) over a moistened filter paper so that every 10 seeds were spread inside each one of the Petri-dishes. The seeds were pre-emergence treated with 1% and 2% of *Thymus capitatus* and *Rosmarinus officinalis*. Simultaneously, another set of dishes was prepared where the seeds inside each dish were soaked in a similar volume

of distilled water (0.0%) to serve as control. All dishes were kept inside an electric incubator at $27^{\circ}\text{C}\pm 2$ for three days. Another batch of the seeds was prepared in the same manner but the time of treatment was 6 days.

At the end of the experimentation, the germination percentage and some growth parameters were taken. Also, the produced seedlings were used immediately in its fresh state where their root apices were detached and used for the cytological preparations. The air dried seedlings were used to determine the nucleic acids content.

The statistical analysis of the obtained data was done according to Snedecor and Cochran, 1989 using the Least Significance Difference test (L.S.D.) at 1% and 5% levels of probability. [9].

Quantitative Estimation of Nucleic Acids: DNA and RNA were extracted according to the method described by Morse and Carter [10]. RNA was estimated colorimetrically by the orcinol reaction as described by Dishe [11] while DNA was estimated by diphenylamine (DPA) color reaction as described by Burton [12].

Cytological Procedure for Mitotic Studies: The roots of the produced seedlings were detached after the end of each period of treatment, washed and fixed in ethanol and acetic acid (3:1 v/v) for 24 hours. Cytological preparations were carried out using Feulgen squash technique [13] and were made permanent by mounting in Canada balsam. Ninety microscopical fields were completely analyzed for each treatment. The photomicrographs were taken from the prepared permanent slides.

RESULTS

Changes in Germination Percentage: Results recorded in Table (1) revealed that the germination percentage was increased by 2.27% in response to pre-emergence treatment of seeds with the lowest concentration of *Thymus* (1%) for 3 days. On the other hand, at the highest concentration applied (2%) the germination percentage was highly significantly reduced by 1.14%. In case of *Rosmarinus*, the germination percentages of the seeds in both treatments were highly significantly reduced by 18.18% and 15.91% at the 1% and 2% treatments for three days, respectively. On the other side, the germination percentage of both seeds treated with *Thymus* and *Rosmarinus* at six days showed highly reduction with the both treatments 1% and 2%.

Changes in Some Growth Parameters: Table (2) showed the results of root length, shoot lengths, fresh weight and dry weights of the seedlings produced from seeds treated with *Thymus* and *Rosmarinus* extracts for three and six days. It is obvious that root length of seedlings treated with 1% and or 2% *Thymus* extract for three and six days showed highly significant increases by 18.57% and 8.44%, respectively for three days and 18.71% and 5.88%, respectively for six days.

On the other hand, the root length of the seedlings treated with 1% *Rosmarinus* extract for three days was increased by 4.64%, while treatment with 2% showed reduction by 40.08%. Treatment for six days showed a reduction in the root length in both 1% and 2% concentrations.

The results of shoot length showed highly significant increases in seedlings treated with 1% and 2% of *Thymus* extract for three and six days. On the other hand, the shoot length of seedlings treated with *Rosmarinus* extract showed a high reduction especially at treatment for six days.

The Fresh weight and dry weights of seedlings treated with *Thymus* extract showed a highly significant increase at 1% and 2% for three days and a highly significant reduction at 2% for six days.

Also the results of fresh weight and dry weights of the seedlings showed a highly significant reduction in all treatments with *Rosmarinus* extract., while the fresh weight of 1% treatment for six days showed a highly significant increase by 24.45%.

Changes in Nucleic Acids Content

After 3 Days of Treatment: It is apparent from Table (3) that DNA content of root tips of seedlings was shown to be either highly significantly increased or non significant changed in response to applying 1% and or 2% of *Thymus* respectively. RNA content of seedlings was shown to be highly significantly increased on using both treatments of *Thymus*.

As a consequence of applying *Rosmarinus*, the content of DNA of root tips of the seedlings was shown to be highly significantly increased with 1%, whereas the same content was highly significantly reduced on using 2%. RNA content of seedlings underwent a pattern of change similar to the corresponding ones in RNA content of seedlings treated with *Thymus*.

After 6 Days of Treatment: The content of DNA in root tips of seedlings was found to be increased either highly significantly or non-significantly in response to 1% and

Table 1: Changes in germination percentage of *Pisum sativum* seeds in response to their pre-emergence treatment with the different concentrations of *Thymus capitatus* L. and *Rosmarinus officinalis* L. for different periods of treatment. Ten replicates were used; each consisted of 10 seeds

Treatments	Concentration	3 days	6 days
Control (H ₂ O)	0.0	88	98
<i>Thymus capitatus</i>	1%	90 +HS	78 -HS
	2%	78 -HS	66 -HS
L.S.D. at 5% level		0.65	1.63
L.S.D. at 1% level		0.93	2.35
<i>Rosmarinus officinalis</i>	0.0	88	98
	1%	72 -HS	68 -HS
	2%	74 -HS	63 -HS
L.S.D. at 5% level		0.88	1.91
L.S.D. at 1% level		1.27	2.75

HS = Highly significant change. S = Significant change. NS = Non-significant change

Table 2: Growth parameters of the seedlings produced from the germinated seeds of *Pisum sativum* in response to their pre-emergence treatment with different concentrations of *Thymus capitatus* L. and *Rosmarinus officinalis* L. for different periods of treatment

Periods of treatment	Treatments	Conc.	Root lengths	Shoot length	Fresh weight	Dry weight
3 days	Control (H ₂ O)	0.0	2.37	0.59	4.50	1.10
	<i>Thymus capitatus</i>	1%	2.81+HS	0.68+HS	4.80+HS	1.40+HS
		2%	2.57+HS	0.98+HS	5.30+HS	1.10NS
	L.S.D. at 5% level		0.022	0.021	0.041	0.018
	L.S.D. at 1% level		0.032	0.023	0.059	0.025
	<i>Rosmarinus officinalis</i>	0.0	2.37	0.59	4.50	1.10
1%		2.48+HS	0.62+HS	3.70-HS	0.80-HS	
2%		1.42-HS	0.45-HS	2.00-HS	0.60-HS	
L.S.D. at 5% level			0.059	0.009	0.129	0.025
L.S.D. at 1% level			0.085	0.013	0.186	0.037
6 days		Control (H ₂ O)	0.0	3.91	2.86	4.50
	<i>Thymus capitatus</i>	1%	4.81+HS	2.96+HS	5.50+HS	1.41NS
		2%	4.14+HS	2.89+HS	3.50-HS	0.70-HS
	L.S.D. at 5% level		0.047	0.005	0.101	0.042
	L.S.D. at 1% level		0.068	0.007	0.145	0.060
	<i>Rosmarinus officinalis</i>	0.0	3.91	2.86	4.50	1.44
		1%	2.18-HS	1.41-HS	5.60+HS	1.35-HS
		2%	2.42-HS	1.37-HS	3.50-HS	0.77-HS
	L.S.D. at 5% level		0.095	0.086	0.106	0.037
	L.S.D. at 1% level		0.136	0.123	0.153	0.053

HS = Highly significant change. S = Significant change. NS = Non-significant change

Table 3: Changes in DNA and RNA contents of root tips of the seedlings produced from the germinated seeds of *Pisum sativum* in response to their pre-emergence treatment with the different concentrations of *Thymus capitatus* L. and *Rosmarinus officinalis* L. for different periods of treatment. Values listed are expressed as µg nucleic acid/g air dried weight. Each value is a mean of three determinations

Treatments	Conc.	3 days		6 days	
		DNA	RNA	DNA	RNA
Control (H ₂ O)	0.0	125.69	248.68	129.79	254.12
<i>Thymus capitatus</i>	1%	135.42+HS	258.06+HS	142.21+HS	279.66+HS
	2%	125.54 NS	246.93+HS	130.09 NS	257.04+HS
L.S.D. at 5% level		0.57	0.60	0.76	1.41
L.S.D. at 1% level		0.82	0.90	1.03	2.03
<i>Rosmarinus officinalis</i>	0.0	125.69	248.68	129.79	254.12
	1%	129.44+HS	273.06+HS	103.72-HS	191.25-HS
	2%	123.09-HS	263.29+HS	88.04 -HS	159.26-HS
L.S.D. at 5% level		0.32	1.24	2.13	4.88
L.S.D. at 1% level		0.46	1.78	3.07	7.01

HS = Highly significant change. S = Significant change. NS = Non-significant change

Table 4: Frequency of mitotic phases, percentage of abnormal mitotic phases, mitotic index and total percentage of mitosis in root tips of the seedlings produced from the germinated seeds of *Pisum sativum* in response to their pre-emergence treatment with different concentrations of *Thymus capitatus* L. and *Rosmarinus officinalis* L. for different periods of treatment

Periods of treatment	Treatments	Conc.	Frequency of mitotic phases			% of abnormal mitotic phases			MI	Total% of mitotic abn.	
			Prophase	Metaphase	Ana-telophase	Prophase	Metaphase	Ana-telophase			
3 days	Control (H ₂ O)	0.0	44.76	20.40	34.84	0.00	0.00	0.00	6.19	0.00	
	<i>Thymus capitatus</i>	1%	25.00	33.57	41.43	0.00	17.02	4.02	7.34+HS	7.38+HS	
		2%	19.55	33.52	46.93	0.00	33.33	14.29	6.51+HS	17.88+HS	
	L.S.D. at 5% level									0.06	0.91
	L.S.D. at 1% level									0.09	1.31
	<i>Rosmarinus officinalis</i>	0.0	44.76	20.40	34.84	0.00	0.00	0.00	6.19	0.00	
		1%	37.56	29.10	33.33	3.31	19.66	14.18	6.94+HS	11.69+HS	
		2%	42.14	26.41	31.45	9.86	31.46	30.19	5.91+HS	21.96+HS	
		L.S.D. at 5% level									0.05
	L.S.D. at 1% level									0.08	1.60
6 days	Control (H ₂ O)	0.0	43.86	23.39	32.75	0.00	0.00	0.00	6.10	0.00	
	<i>Thymus capitatus</i>	1%	25.93	31.62	42.45	12.09	11.72	15.44	6.66+HS	13.39+HS	
		2%	16.86	38.66	44.48	5.17	9.02	15.03	6.12 NS	11.05+HS	
	L.S.D. at 5% level									0.03	0.72
	L.S.D. at 1% level									0.05	1.04
	<i>Rosmarinus officinalis</i>	0.0	43.86	23.39	32.75	0.00	0.00	0.00	6.10	0.00	
		1%	33.96	27.92	38.11	20.00	13.51	18.81	4.77+HS	17.74+HS	
		2%	32.08	22.64	45.28	0.00	14.58	7.29	4.22+HS	6.60+HS	
		L.S.D. at 5% level									0.10
	L.S.D. at 1% level									0.14	1.30

HS = Highly significant change. NS = Non-significant change

Table 5: Types and frequency of abnormal mitotic phases in root tips of the seedlings produced from the germinated seeds of *Pisum sativum* in response to their pre-emergence treatment with different concentrations of *Thymus capitatus* L. and *Rosmarinus officinalis* L. for different periods of treatment

Periods of treatment	Treatments	Conc.	% of Prophase abnormalities		% of Metaphase abnormalities		% of Ana-telophase abnormalities		% of Interphase abnorm.
			Dist.	Dist.	C-meta.	Lagg.	Dist.	Lagg.	
3 days	Control (H ₂ O)	0.0	-	-	-	-	-	-	-
	<i>Thymus capitatus</i>	1%	-	15.60	-	1.42	2.87	1.15	-
		2%	-	28.33	2.5	2.5	10.71	1.79	1.79
	<i>Rosmarinus officinalis</i>	0.0	-	-	-	-	-	-	-
		1%	3.31	13.68	1.70	4.27	10.45	-	3.73
		2%	9.86	16.85	6.74	7.87	13.21	12.26	4.72
6 days	Control (H ₂ O)	0.0	-	-	-	-	-	-	-
	<i>Thymus capitatus</i>	1%	12.09	5.41	-	6.31	4.03	3.36	8.05
		2%	5.17	6.02	-	3.01	10.46	1.31	3.27
	<i>Rosmarinus officinalis</i>	0.0	-	-	-	-	-	-	-
		1%	20.00	8.11	1.35	4.05	3.96	6.93	7.92
		2%	-	14.58	-	-	7.29	-	-

or 2% of *Thymus*, respectively. RNA content of seedlings was shown to be highly significantly increased with all concentrations applied.

Due to application of *Rosmarinus*, the contents of DNA in root tips of seedlings were shown to be highly significantly reduced with all concentrations applied. RNA content of seedlings underwent a pattern of changes as that of the corresponding changes in DNA content.

The magnitude of such induced reductions in DNA and RNA contents of seedlings by all concentrations of *Rosmarinus* were shown to be sharply higher than that of the corresponding reductions in the same contents of *Thymus*.

Changes in Mitotic Activity:

After 3 Days of Treatment: Data presented in Table (4) revealed that the frequencies of different mitotic phases of seedlings followed a similar sequence of change in response to applying various concentrations of *Thymus*. Whereas the frequency of prophase was shown to be inconsistently reduced as the concentration applied was increased, those of metaphase and ana-telophase were shown to be reversely changed. Mitotic index (MI) of seedlings was shown to be highly significantly increased on using 1% and/or 2% of *Thymus*.

Applying various concentrations of *Thymus* did not induce abnormal prophase in both 1% and 2% treatments, whereas the same treatments were shown to induce

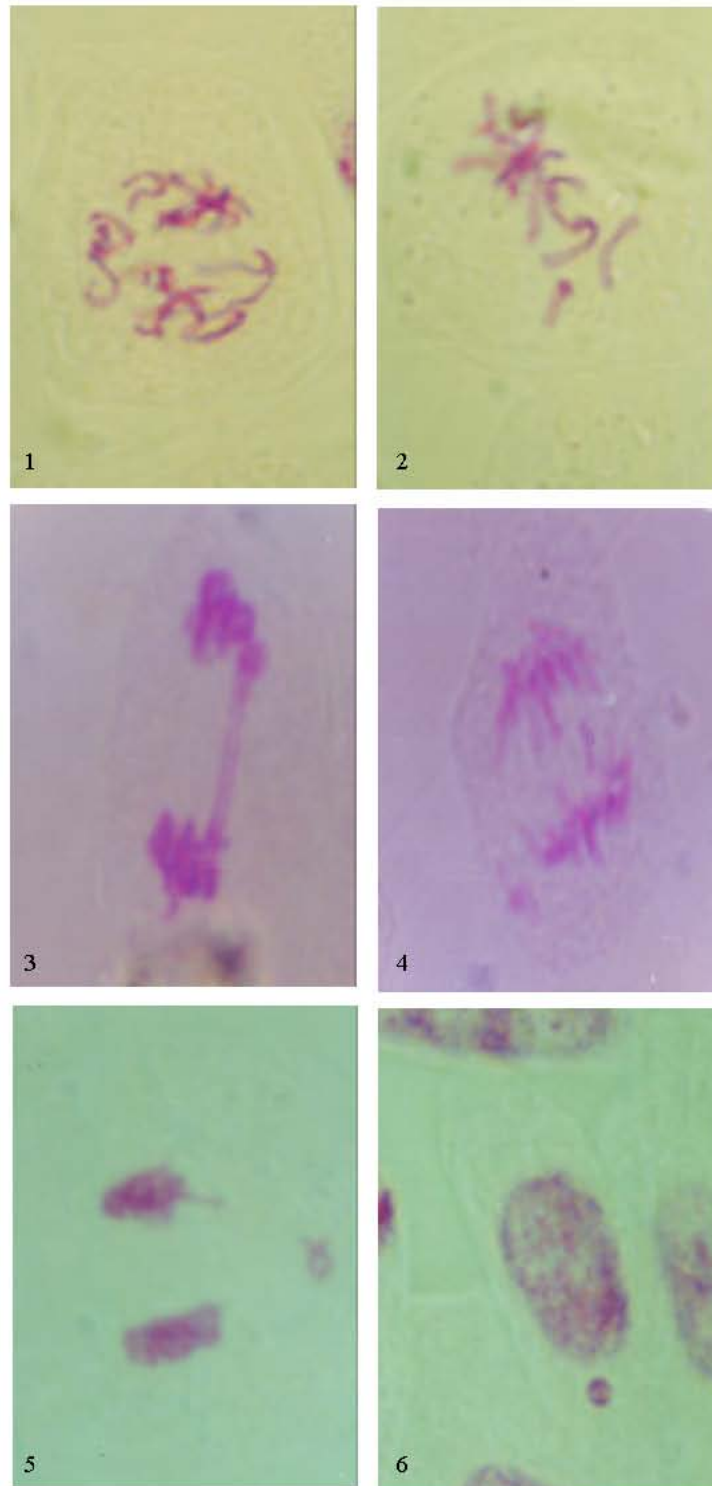


Plate (1): Mitotic abnormalities in root tip cells of *Pisum sativum* seedlings in response to their pre-emergence treatment with different concentrations of *Thymus capitatus* L. and *Rosmarinus officinalis* L. for different periods of treatment. 1. Disturbed Prophase 2. Disturbed metaphase with lagging chromosomes. 3. Ana-telophase with bridges 4. Disturbed Anaphase with lagging chromosomes. 5. Ana-telophase with lagging chromosome. 6. Micronucleus.

metaphase and ana-telophase abnormalities. The percentage of such mitotic abnormalities was shown to be increase with the magnitude of the concentration applied. Moreover, the total percentage of mitotic abnormalities in root tip cells of seedlings exhibited high significant increases on using various concentrations of *Thymus*.

Due to application of *Rosmarinus*, the frequency of prophase in root tip cells of seedlings was shown to be reduced on using 1% and/or 2% of *Rosmarinus* respectively. On the other hand, the frequency of metaphase was increased whereas that of ana-telophase was decreased on using all concentrations applied when being compared with those of the respective controls. MI was highly significantly increased with the lowest concentration used (1%). Whereas it was reversely changed with the higher concentrations applied (2%).

The percentage of abnormal mitotic phases in root tip cells of seedlings was shown to be increase with the magnitude of the concentration applied of *Rosmarinus*. The total percentage of mitotic abnormalities of the seedlings recorded high significant increase with all concentrations applied of *Rosmarinus*.

After 6 Days of Treatment: Whereas the frequency of prophase was reduced and that of metaphase and ana-telophase was increased in seedlings in response to various concentrations applied of *Thymus*. MI of seedlings was increased either highly significantly or non-significantly on using 1% or 2% of *Thymus*. The same index was highly significantly reduced with all concentrations used of *Rosmarinus*.

The percentage of mitotic abnormalities of seedlings treated with *Rosmarinus* varied according to the magnitude of the concentration applied. Moreover, the total percentage of mitotic abnormalities recorded high significant increase in response to various treatments applied.

Several types of mitotic abnormalities were shown to be induced in root tip cells of the seedlings in response to various treatments for three and six days (Table 5 and Plate 1). The most common abnormal mitosis was disturbed, C-metaphase, lagging and bridge. In addition, micronucleated cells were shown to be induced at interphase.

DISCUSSION

It is known that *Thymus capitatus* and *Rosmarinus officinalis* extracts not only used for flavoring foods but also used in food industry and medicine due to the presence of some important antioxidant oil and phenolic

components [14, 15]. It is used also to prevent oxidative degradation of oil and lipid containing foods [16-20].

Lassaad *et al.* [21] mentioned that thyme contains an essential oil endowed with a therapeutic action due to the aromatic alcohols such as thymol and carvacrol which have an antiseptic, antibacterial and analgesic action. Thyme contains the following components thymol, *p*-cymene, carvacrol, pinene, linalool, α -terpinyl acetate, cineole, caryophyllene, geraniol, borneol, α -terpineol, germacrene D, linalyl acetate, nerolidol, camphor, llemol, phenols, sesquiterpene, cadinene and terpinene [22, 23].

Several studies have focused on antimicrobial, antifungal, antioxidant and radical-scavenging properties of essential oils of thyme in order to identify the responsible compounds of these actions [24-27].

Juven *et al.* [28] demonstrated the role of terpene phenols which seem to play an outstanding role in joining the amine and hydroxylamine groups of the proteins of the bacterial membrane altering their permeability and resulting in the death of the bacteria.

Similar results were recorded by Ultee *et al.* [29] and Knowles *et al.* [30] in which they demonstrated that thymol and carvacrol have been considered as a biocidal, resulting in bacterial membrane perturbations that lead to leakage of intracellular ATP and potassium ions and ultimately cell death.

Spasmolytic as well as antioxidant activities [31, 32] were also reported that for the phenolic oil extract of the *Thymus* plant. There is some evidence that minor components play a critical part in biological activities, possibly by producing a synergistic effect between other components.

Alberto *et al.* [33], Atti-Santos *et al.* [34] and Szumny *et al.* [35] demonstrated the chemical composition of the essential oil of the *Rosmarinus officinalis* L. these are categorized into three groups phenolic diterpenes possessing abietic acid framework, flavonoids and phenolic acids. Ozlem *et al.* [36] mentioned that carnosic acid (CA), carnosol, abietane-type diterpenes, rosmarinic acid (RA) and hydroxycinnamic acid ester are the main antioxidant compounds present in rosemary. These compounds, together with other isoprenoids such as sterols, isoprene, mono- and diterpenes, tocopherols or carotenoids play a photoprotective role and are considered as bioactive constituents.

Rasooli *et al.* [37] demonstrated that the essential oils of *Rosmarinus officinalis* could be safely used as preservative materials on some kinds of foods to protect them from toxigenic fungal infections. In which, aflatoxin B1 (AFB1) is a highly toxic and carcinogenic metabolite produced by *Aspergillus* species on food and agricultural

commodities. Natural products may regulate the cellular effects of aflatoxins and evidence suggests that aromatic organic compounds of spices can control the production of aflatoxins. Antifungal activities of the oils were studied with special reference to the inhibition of *Aspergillus parasiticus* growth and aflatoxin production.

Most of the Lamiaceae plants contain considerable amounts of rosmarinic acid, which, in addition to well known free radical scavenger properties and a natural antioxidant which is found as a secondary metabolite in rosemary (*Rosmarinus officinalis*) displays antibacterial and antifungal properties [38, 39].

Antimicrobial and antifungal properties of rosmarinic acid against 40 different strains belonging to 10 genera of bacteria and 25 different strains belonging to 25 species of fungi have been published recently [40]. The bacterial strains tested were *Salmonella*, *Serratia*, *Enterobacter*, *Proteus*, *Providentia*, *Echerichia coli*, *Streptococcus*, *Klebsiella* and *Staphylococcus*. In all bacterial strains, 100% inhibition was observed at dosages of 0.5 mg/ml. More detailed study on 29 strains of *Staphylococcus aureus* showed 70% inhibition at concentration of 0.06 mg/ml and 80% at 0.12 mg/ml [40]. Antifungal activity was tested against strains of Adelomycetes, Mucorales, *Rhizopus*, Endomycetes, Plectomycetes and Pyrenomycetes [40, 41].

Carnosol and ursolic acid which are found in *Rosmarinus officinalis* inhibit tumor promotion by inhibiting the Tyrosine kinases and Ornithine decarboxylase activity. Other compounds found in rosemary called diterpenoids, also have antioxidant activity [42]. Carnosol has also shown to reduce nuclear factor-kappaB (NF-kappaB) [43] and the anti-apoptotic protein Bcl-2 [44].

The obtained data from tables Tables (1) and (2) revealed that *Thymus capitatus* and *Rosmarinus officinalis* caused depression in germination percentage and some growth parameters and these results concomitant with the results of nucleic acids content. Also reductions in DNA and RNA contents in root tips of some treated seedlings were found in (Table 3). It indicated that *Thymus* and *Rosmarinus* extracts may trigger a drastic depressive impact on the machinery of nucleic acids synthesis in seedlings. Findings were obtained by [45] were found that DNA-damaging activity of *Bacillus subtilis* due to the estragol component of the oil of *Thymus capitatus* L. and. Also Elizabeth *et al.* [46] found that the commonly used spice and flavoring agent, derived from the leaves of *Rosmarinus officinalis* L., displays antioxidant properties in foods and in biological systems and inhibited DNA adduct formation by 80% and mRNA expression was 50% lower in the presence of

rosemary components. Also Aherne *et al.* [47] demonstrated that rosemary was the most toxic and induced DNA damage.

Table (4) showed that the mitotic activities of root tip meristems of seedlings in response to various treatments have differential responses. Whereas the number of cells entering mitosis in seedlings was shown to be mostly markedly increased. The stimulated mitotic activity of seedlings could be described to the enhanced duration of the cell cycle and stimulation of DNA-protein synthesizing machinery whereas the inhibited mitotic activity seedlings might be attributed to blocking and/or prolonging duration of mitotic cycle and depressed DNA-protein synthesizing machinery in response to various treatments. [48-50].

Table (5) showed the high percentage of chromosomal abnormalities induced in root tip cells meristems of seedlings coupled with the appearance of micronucleated cells as a consequence of various spices treatments pinpoint to the drastic impact of spices on the chromosomal structure and/or their behavior via depression of regular dissolution of nuclear membrane at prophase, disturbance in the formation and function of chromosomal movement-mechanism at metaphase and ana-telophase, liquefaction of DNA, as well as enhancement of occurrence of certain chemical or physical events along DNA molecule which produce high amount of energy causing micronuclei at interphase. Similar chromosomal abnormalities were recorded by other investigators such as Sengupta and Ghosh, [51] and Nyarai-Horvath *et al.* [52].

Despite of *Thymus capitatus* and *Rosmarinus officinalis* are used for flavoring foods, food industry, antimicrobial, antifungal and antioxidant agents due to the presence of some important phenolic components in their essential oils, the obtained results showed significant reductions in germination percentage, some morphological features, nucleic acids content, and mitotic index and also the appearance of high percentage of chromosomal abnormalities. The present work demonstrated the bad effect of these spices when used in large quantities and this may be due to the poisonous effect of aromatic compound including phenolic compounds and terpene-phenolic derivatives which are poisonous and can cause painful rash. This conclusion is in consistence with those offered by Houlihan *et al.*, [53] and Imad *et al.* [54]. Also Huang *et al.*, [55], Calabrese *et al.*, [56] and Moss *et al.*, [57] demonstrated that rosemary essential oil is potentially toxic if ingested. Large quantities of rosemary leaves can cause adverse reactions, such as coma, spasm, vomiting, and pulmonary edema that can be fatal.

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