The antibacterial effect assay of a few officinal plants

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Abstract
From very old times plants were used for man or animal health, with good results in slightly, functional injuries or in incipient stages of diseases. In chronic, when already appeared organic affection, officinal plants has an adjuvant part and can contributes to a partial reversibility of symptoms or lesions. This study was been performed using aqueous solutions or essential oils from *Mentha spicata, Ocimum basilicum, Thymus vulgaris* and *Rosa sp.* with bacterial cultures on usual or special culture mediums of Bacillus cereus, Escherichia coli, Pseudomonas aeruginosa, Salmonella enterica and Staphylococcus aureus. In view of antibacterial assay emphasizing were been used small pieces of absorbent paper immersed in solutions or essential oils above mentioned officinal plants. These pieces of papers then were been deposed in usual and special culture mediums containing the mentioned species of bacteria and then incubated 24 hours. The obtained result emphasized by the inhibition areas development recommend the antibacterial effect of essential oils of *Rosa sp.* (25.5 mm inhibition area), *Mentha spicata* (16.5 mm), *Thymus vulgaris* (7.3 mm) and *Ocimum basilicum* (5.1 mm) in therapeutic purpose, both in intern and extern way in infections caused by Bacillus cereus, Escherichia coli, Salmonella enterica and Staphylococcus aureus, in incipient stages.

Keywords: antibacterial assay, bacterial culture mediums, essential oils, inhibition areas.

1. Introduction

From very old times man tried to find remedies to treat painful diseases for him and his animals (3, 6). All accumulated knowledge were been handed down from generation to another. In that period men cohabited in harmony with nature, being more interested by it preservation. It is possible that in ancient times men were based in remedies selection, like diseased animals does, on instinct, but many times they used empiric methods, trying and observing their effects (2). In almost all world people the plants therapeutic effects were been considered supernatural, and healing was the exclusive privilege of the warlocks. In our times it is well known the action of officinal plants of human and animal body through phytotherapy, a new science with an efficiency proved from very ancient times (3). The treatment with officinal plants has good results in incipient stages of diseases, while in chronic diseases the role of phytotherapy becomes adjuvant, contributing to a partial reversibility of symptoms and lesions (1, 6).

This study aimed to emphasize the antibacterial effects of a few active substances from some plants structure using bacterial species implicated in appearance and evolution of some pathological processes in man and animals.

2. Materials and methods

The experimental method used aqueous solutions and essential oils of the following officinal plants: Mentha spicata, Ocimum basilicum, Thymus vulgaris and Rosa sp. Were been transplanted on

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fresh aqueous culture mediums the following species: Bacillus cereus, Escherichia coli, Pseudomonas aeruginosa, Salmonella enterica and Staphylococcus aureus. Escherichia coli was also transplanted on Levin special culture medium and Salmonella enterica on Simmons special culture medium. Then were been prepared the above mentioned officinal plants, in the view of antibacterial effect assay performing on transplanted bacteria. In Berzelius glasses were been deposed 100 gram dry above mentioned officinal plants with 10 ml distilled water, in each glass and then were well mixed. After 24 hours from each Berzelius glass were been collected plants with a spatula on Petri plates and in each aqueous solution of plants being incorporated small round pieces of filter paper. In the same time were been impregnated round pieces of filter paper in essential oils of Mentha spicata, Ocimum basilicum, Thymus vulgaris and Rosa sp. After 16-18 hours, while the bacterial culture mediums were been held in thermostate at 37 Celsius degree were been prepared 10 Petri plates with nutrient agar, Levin special medium and Simmons special medium. Were been inoculated two plates with each of above mentioned bacteria species. On five Petri plates were been put small round pieces of filter paper impregnated with aqueous solutions officinal used plants and on five plates were been put pieces of filter paper impregnated in essential oils of used officinal plants (table 1).

All Petri plates were been than incubated 24 hours at 37 Celsius degree. After 24 hours of incubation the obtained results emphasized through inhibition area, around each filter paper round piece are carefully measured with a ruler and a magnifying glass.

3. Results and discussion

In the case of Petri plates with Escherichia coli, Bacillus cereus, Pseudomonas aeruginosa, Salmonella enterica and Staphylococcus aureus on nutrient agar culture medium was been observed that around pieces of filter paper impregnated with aqueous solutions of Mentha spicata, Ocimum basilicum, Thymus vulgaris and Rosa sp. Were not developed inhibition areas, only a few, with small dimensions around filter paper impregnated with Rosa sp. in the Petri plate with Bacillus cereus (0.1 mm) and Salmonella enterica (0.3 mm) (Table 2).

The explanation may be that these solutions of 100 grams of plants in 50 ml distilled water didn't contained active substances in sufficient quantities to emphasize an antibacterial effect, with above mentioned two exceptions. In the case of round filter paper pieces impregnated in officinal plants essential oils were been obtained negative results on nutrient agar with Pseudomonas aeruginosa and the Simmons medium with Salmonella enterica. The most reduced value of inhibition area was been obtained in the case of Salmonella enterica (0.2 mm). The most increased values were been obtained in the case of Bacillus cereus around the paper impregnated with essential oil of Rosa sp. (9 mm), Mentha spicata (8 mm) and in the case of Bacillus cereus, around impregnated paper with Rosa sp. Essential oil (7 mm) (Table 3). Regarding to amount of inhibition areas diameters dimensions obtained in the case of essential oils was been observed that the most increased value was been obtained at Rosa sp. (25.5 mm), Mentha spicata (16.5 mm), Thymus vulgaris (7.3 mm) and Ocimum basilicum (5.1 mm). Taking into discussion the amount of inhibition areas diameters calculated on bacteria species, the most increased value was been obtained in the case of Bacillus cereus (19.5 mm), Escherichia coli on Levin medium (11.3 mm), Salmonella enterica on nutrient agar (10.7 mm), Escherichia coli on nutrient agar (7 mm) and Staphylococcus aureus (5.9 mm).

Table 1. The researching material preparation for antibacterial effect assay

Petri plate	Species of bacteria	Mentha spicata		Ocimum basilicum		Thymus vulgaris		Rosa sp.	
		Plant	Oil	Plant	Oil	Plant	Oil	Plant	Oil
1.	Bacillus cereus	+	+	+	+	+	+	+	+
2.	Escherichia coli	+	+	+	+	+	+	+	+
3.	Pseudomonas aeruginosa	+	+	+	+	+	+	+	+
4.	Salmonella enterica	+	+	+	+	+	+	+	+
5.	Staphylococcus aureus	+	+	+	+	+	+	+	+

Table 2. The obtained results in the case of officinal plants aqueous solutions

Petri plate	Species of bacteria	Mentha spicata (Ø of inhibition area)	Ocimum basilicum (Ø of inhibition area)	Thymus vulgaris (Ø of inhibition are)	Rosa sp. (Ø of inhibition area)
1.	Escherichia coli (nutrient agar)	0 mm	0 mm	0 mm	0 mm
2.	Bacillus cereus	0 mm	0 mm	0 mm	0.1 mm
3.	Pseudomonas aeruginosa	0 mm	0 mm	0 mm	0 mm
4.	Salmonella enterica (nutrient agar)	0 mm	0 mm	0 mm	0.3 mm
5.	Staphylococcus aureus	0 mm	0 mm	0 mm	0 mm
6.	Salmonella enterica (Simmons)	0 mm	0 mm	0 mm	0 mm
7.	Echerichia coli (Levin)	0 mm	0 mm	0 mm	0 mm

Table 3. The obtained results in the case of officinal plants essential oils

Petri plate	Species of bacteria	Mentha spicata (Ø of inhibition area)	Ocimum basilicum (Ø of inhibition area)	Thymus vulgaris (Ø of inhibition are)	Rosa sp. (Ø of inhibition area)	Total
1.	Escherichia coli (nutrient agar)	1.5 mm	1.9 mm	1.1 mm	2.5 mm	7 mm
2.	Bacillus cereus	8 mm	0 mm	2.5 mm	9 mm	19.5 mm
3.	Pseudomonas aeruginosa	0 mm	0 mm	0 mm	0 mm	0 mm
4.	Salmonella enterica (nutrient agar)	3 mm	0.5 mm	0.2 mm	7 mm	10.7 mm
5.	Staphylococcus aureus	1 mm	0.4 mm	1.5 mm	3 mm	5.9 mm
6.	Salmonella enterica (Simmons)	0 mm	0 mm	0 mm	0 mm	0 mm
7.	Echerichia coli (Levin)	3 mm	2.3 mm	2 mm	4 mm	11.3 mm
	Total	16.5 mm	5.1 mm	7.3 mm	25.5 mm	

The best results in this experiment were been obtained in the case of essential oils, in comparison with aqueous solutions.

The explanation is that officinal plants in aqueous extracts was used a quantity of 100 grams from each plant, while in essential oils the quantity is 5-8 kg plants for one milliliter of essential oils, like some authors sustained (2). In such a big quantity of plants are concentrated a big quantity of active substances from plants and the antibacterial effects was easy emphasized, like many authors observed in their scientific papers (4, 5, 6).

4. Conclusions

In the case of used officinal plants aqueous extracts in the experiment, in majority the obtained results were been negatives. These results prove that the aqueous solutions of Mentha spicata, Ocimum basilicum, Thymus vulgaris and Rosa sp. contain insufficient active substances to emphasize antibacterial effects. Taking into consideration the positive obtained results in the case of essential oils from the same plants is recommended that these oils can be applied in therapeutic purpose in infections with Bacillus

cereu, Escherichia coli, Salmonella enterica, Staphylococcus aureus, only in incipient stages. The application of officinal plants extracts in therapy represents a healthily and efficient alternative of antibiotics, without the rescue of resistance phenomena appearance, of equilibrium in intestinal flora disturbance or another negative effects on man and animal health, with the condition of their application in incipient stages of diseases.

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