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Evaluation of the Antibacterial Effect of Essential Oil of *Cuminum cyminum L*

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ABSTRACT

The attention focused on herbs and spices such treatment source. This study contributed to the knowledge of the antimicrobial potential of *Cuminum cyminum L* diluted and pure. The essential oil was extracted from the dried fruit of the plant by steam distillation.
The antibacterial activity of the essential oil of *Cuminum cyminum L* was determined by aromatogram through two methods: the diffusion method on agar medium, technical records and wells, and the dilution method in liquid medium on such pathogenic bacteria Salmonella thyphi, Vibrio cholerae, Shigella spp, Staphylococcus aureus and Clostridium sp, and beneficial bacteria, probiotic Escherichia coli, Streptococcus sp, Lactobacillus sp. The yield was about 3.54% in the essential oil. The ET has been active on all bacteria tested Gram negative and Gram positive with a predominant sensitivity of Gram-negative bacteria. Effective action on bacterial pathogens was demonstrated as shown in the case of *Staphylococcus aureus*, *Vibrio cholerae* and *Salmonella thyphi* and *Shigella sp*. Nevertheless no effect on beneficial bacteria such as *Lactobacillus sp* which is very durable. Cumin can be used as a prebiotic and antimicrobial agent.
KEYWORDS: Essential oil, *Cuminum cyminum*, antibacterial activity.

1- INTRODUCTION

Medicinal plants still remain the first reservoir of new drugs having biological properties which find many applications. This renewed interest is firstly the fact that these plants represent an inexhaustible source of bioactive substances, and also the emergence of bacterial strains resistant to antibiotics [1]. Faced with this problem and as part of the development of natural resources, the objective of this study was focused on the identification of essential oil antimicrobial potential of the Cuminum cyminum The in vitro on different bacterial species and pathogens else usually commensal gut.

2- MATERIAL AND METHODS

2.1 Plant Material

Cumin (*Cuminum cyminum*) plant is a thin, hairless, and annual herbaceous, with a height of 20-60 cm. The dried seeds (fruit) of a pale green color, elliptical shape and do not the length is between 5 and 6 mm. In herbal medicine, the seeds are used in their natural state (infusion or decoction).

2.2 Bacteria used

The choice of the bacteria was carried on frequent strains in human pathology and other beneficial bacteria or probiotics play a key role in the balance of our intestinal microbiota. Gram positive bacteria like *Staphylococcus aureus, Streptococcus sp, Clostridium sp; Lactobacillus sp* and were tested and Gram negative *Shigella sp*, *Escherichia coli, Salmonella thyphi* and *Vibrio cholera*

2.3 Extraction of the essential oil by steam distillation

In a flask, introduce 100 g of cumin seeds added to 500 ml of distilled water, the whole was brought to a boil by the heating mantle for 2 hours and 30 min at 95 ° C [2].

The boiling of the mixture creates a water vapor stream of charged chemical compounds. The condensation of these vapors is it possible to obtain a distillate to be treated, by an organic solvent (cyclohexane) allowing the separation of two phases: an aqueous and the other organic. The latter was placed in a rotary evaporator to evaporate the solvent (cyclohexane) at a temperature of 70 ° C and recovering the pure essential

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oil. The latter was collected in glass vials and shaded setting at a temperature of 4 ° C to avoid any deterioration or reaction that can alter its composition [3]

2.4 Determination of performance

The yield of essential oil (TEA) was defined as the ratio between the mass of the oil obtained after extraction (M ') and the mass of the plant material used (M) .The yield was expressed as a percentage calculated using the following formula [4].

$$RHE = \frac{M'}{M} X\ 100$$

RHE: essential oil yield of cumin%.
M: mass of the essential oil obtained by gram.
M: mass of seeds used in grams.

2.5 organoleptic characterization of the essential oil of cumin

Essential oils must meet analytical characteristics established by national and international commissions of experts.Pour know the quality of the essential oil of cumin study was conducted organoleptic tests (color, odor and appearance) and compare with those of the AFNOR standard 1990 [5].

2.6 Study of antibacterial activity HE of *Cuminum Cyminum*

The antibacterial activity of the essential oil of Cyminum cuminum was evaluated by the agar diffusion method. The purity and identity of the bacterial strains were confirmed by conventional biochemical tests. From a pure culture of 18- 24 hours on non-selective medium adapted to the requirements of each strain, the inoculum suspension in sterile saline equivalent to McFarland standard 0.5 ~ 10 8 CFU / ml was performed for each strain. This inoculum was inoculated by flooding on Petri plates containing Mueller-Hinton agar [6]. Disks impregnated by the various concentrations of pure and diluted ET were gently deposited on the surface of the agar. The Petri dishes were first left for 30 minutes to a pre-diffusion of oil before being incubated at 37 ° C for 24 h. [7] [8]. All tests are made Duplicate.

3. RESULTS

3.1 Essential Oil yield

Calculating yields has not only to assess the essential oils extracted from a plant, but also to consider the amount of seeds to be taken in case of need for any similar study; which would make the rational and sustainable use of so-target species. For our sample, the yield of essential oil was of the order of 3.53%.

3.2 Tasting essential oil cumin

After extraction, the organoleptic characteristics of our essential oil were examined and compared with those of the AFNOR standard [5].This analysis indicates that no significant difference is mentioned. The results are shown in Table 01.

Table 01: Essential oil organoleptic characters cumin

Organoleptic characters	Color	Apparance	Smell
Essential oil	dark yellow	Liquid	Characteristic odor, Aromatic, spicy
A.F.N.O.R.	dark yellow	Liquid	Fatty, Characteristic odor, Aromatic,

3.3. Evaluation of antibacterial activity

3.3.1 aromatogram

From the set of preliminary tests, pure EH dose of cumin was chosen is 10µl, or the effect began to be significant, with differences between the diameters of zones of inhibition of bacteria tested (Fig 01).

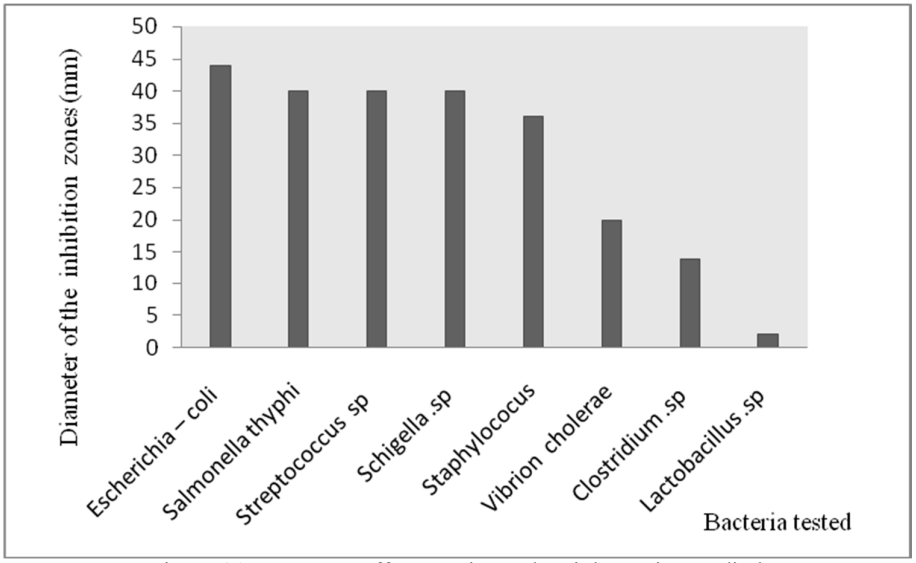

Figure 01 : Pure HE Effect cumin on the eight strains studied

This histogram summarized the results of the general aromatogram of all studied bacteria or was deduced significant sensitivity *Escherichea coli* overlooked the 10µl dose ET cumin with a diameter of 44mm, followed by *Salmonella thyphi*, *Streptococcus sp* and *Shigella sp* 40mm; *Staphylococcus aureus* has a diameter of 36 mm, *Vibrio cholera* 20mm; *Clostridium sp* 14 mm. However, it was observed a resistance of *Lactobacillus sp.* diameter having a 2 mm only.

Regarding diluted doses, we opted for binary dilutions: the first dilution (D1) applied corresponds to 0.9 x 103mg / ml, the second (D2) 0.45 x 103mg / ml and the third (D3) 0.225 x 103mg / ml. A Witness: dimethyl sulfoxide (DMSO).

The species Staphylococcus aureus, the zones of inhibition were identified whose diameters are respectively 36 mm for ET pure and 2mm for the (D1 = 0.9 x 103mg / ml). However no area is observed for other dilutions and the witness. The results of the effect of pure and diluted ET have been illustrated in (Fig 02).

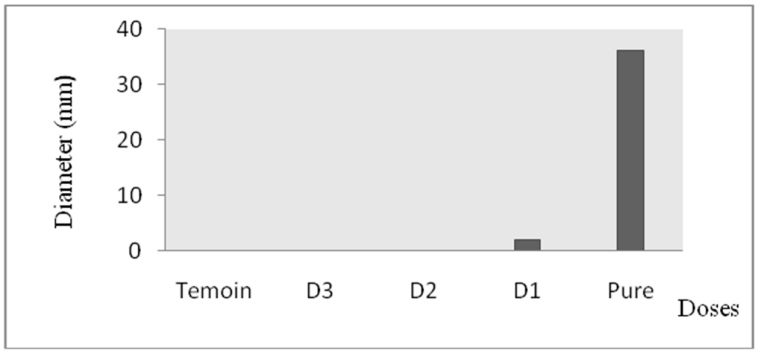

Figure 02: Aromatogram *Staphylococcus aureus*

Our results are consistent with the work of [9] that confirm the effect of cumin oil on Staphylococcus aureus. Other authors [10] showed that the synergistic effect between the HE of *Cuminum cyminum* and a probiotic has a very powerful inhibitory effect on *Staphylococcus aureus.*

Streptococcus sp strain was very sensitive to HE pure by a 40 mm zone of inhibition diameter of 10mm (D1) and (D2), a 6mm diameter (D3) with an absence of area inhibition for the control (DMSO). (Fig 03).

Figure 03 : Aromatogram *Streptococcus sp*

Our results correlate with another study that confirms the antibacterial effect of HE cumin on *Streptococcus sp*. that don't inhibit diameter 54 mm. [11].

In the presence of the same pure dose of 10µl, *Clostridium sp* strain was less sensitive than the other strains, having a 14 mm zone of inhibition. Dilution (D1) gave a diameter of 2mm. However, other dilutions showed no effect. (Fig 04).

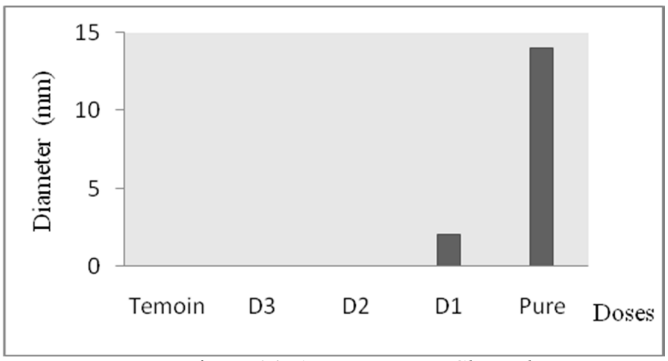

Figure 04: Aromatogram *Clostridium sp*

Our results for Clostridium .sp are consistent with the work of [12] which confirmed the antibacterial effect of cumin ET on that strain.

Furthermore, *Lactobacillus sp* presented vis-a-vis resistance pure and diluted HE (Fig 05). This kind of bacterium seems to multiply in the presence of the active ingredients of HE cumin. Many studies indicate the use of the genus Lactobacillus as a probiotic in combination with ET cumin against pathogens [13].

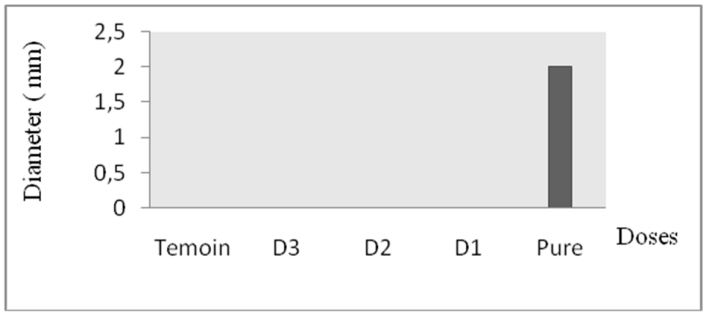

Figure 05 : Aromatogram *Lactobacillus .sp*

For *Escherichia coli,* Gram-negative bacteria, the application of pure HE presented a significant effect in a 44 mm zone of inhibition diameter, the (D1) and (D2) had respective diameters of 6mm and 4mm. No zone of inhibition to (D3) and DMSO (Fig 06).

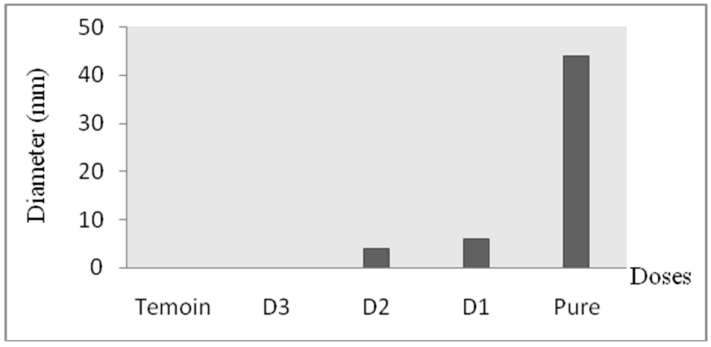

Figure 06: Aromatogram *Escherichia – coli*

Our results are in agreement with the works and Nisha Chaudhary et al. [12] which confirm the antibacterial effect of HE cumin *Escherichia coli*. Another study [11] which shows a strong inhibitory effect of HE cumin on. *Escherichia coli.* whose inhibition diameter reaches 60 mm.

Likewise, for *Salmonella thyphi* showed sensitivity to ET pure with an area of ihnibition 40mm. A 10 mm diameter (D1) and (D2); 6mm (D3) with no area for the control (DMSO). (Fig 07). Our result is consistent with other studies that have found that HE cumin has an effect on *Salmonella thyphi* [12]

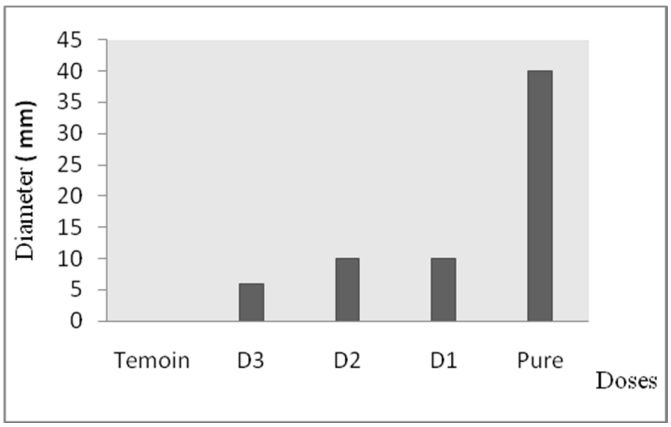

Figure 07 : Aromatogram *Salmonella thyphi*

Shigella sp has been sensitive vis-à-vis the pure HE the cumin with a diameter of 40 mm, but for diluted HE D1 and D2, the mentioned diameters are 6mm and 8mm respectively and no areas for (D3) and the control (Fig 08).

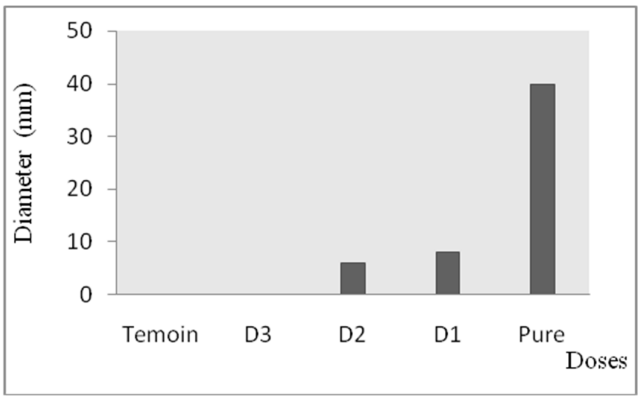

Figure 08 : Aromatogramme de *Shigella .sp*

The pathogenic strain, *Vibrio cholerae*, presented a diameter of 21 mm and 10mm HE pure D1. A lack of areas inhibition was noted for the rest of dilutions (D2 ; D3) and the control (Fig 09).

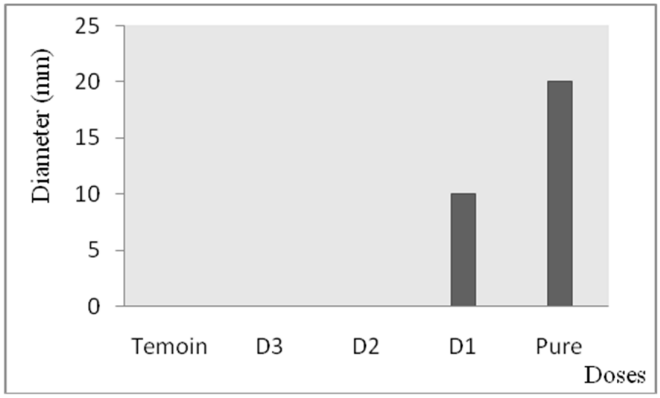

Figure 09 : Aromatogram *Vibrion cholerae*

4. DISCUSSION

The results of our study showed that the essential oil of Cuminum cyminum, whatever pure or diluted wore an antibacterial effect on the majority of bacteria tested.

These results correlate with the literature, according to (13) the sensitivity of a microorganism with essential oils not only depends on the properties of essential oil but also the nature of the microorganism itself (morphology, structure, metabolic type, nutritional, pathogenicity ...).

In our study, the mode of action is not clearly understood given the nature, diversity and complexicité molecules present in the essential oil. The activity of a plant substance depends on several factors including the method of extraction and concentration of active ingredients.

Besides the diversity of bacterial cells, their nature, their different trophic types their particular metabolism , the antibacterial activity appears to result from a combination of several modes of action. [13] The hydrophobicity of certain molecules in essential oils, allows them to be dissolved in the membranes, which causes a destabilization of the structure and an increase in membrane permeability [14]. These changes cause an ion leakage of intracellular compounds and resulting cell death [15] and [16].

Researchers have tested in vitro, on fifteen bacteria antibacterial activity hydrosols sixteen spices. Hydrosols cumin were active, only Bacillus brevis, Enterobacter aerogenes and Escherichia coli O157: H7. The essential oil of caraway showed stronger antibacterial effect against Escherichia coli, Staphylococcus aureus and Listeria monocytogenes. [17]

This qualitative and quantitative study of the antibacterial effect of HE cumin has been very important because we have obtained promising results, in which some strains appear to be distinguished by a very high sensitivity to others.

The activity of essential oils is often equated with bacteriostatic. However, studies have shown that essential oils can also have bactericidal properties. [18]

5. Conclusion

The traditional use of cumin seeds (Cuminum Cyminum) inspired us to assess the biological properties of the essential oil. In the present work, we tried to contribute to the study of the in vitro antibacterial activity. The review of all of our results, we were applied in drawing a number of conclusion.Le yield of about 3.54% in the essential oil extracted from the seeds of cumin by steam distillation. The qualitative and quantitative study of antibacterial and dilute our pure essential oil has been very interesting because it presented a remarkable potential for inhibiting all strains studied except Lactobacillus bulgaricus. We note also the impact of the HE cumin on Gram positive bacteria and negative bacteria, the latter seems more sensitive. An antibacterial effect on pathogenic strains .sp Shigella, Vibrio cholerae, Salmonella and Staphylococcus aureus has been shown thyphi. The quality, efficiency and economy of obtaining the essential oil depends not only on the origin and characteristics of the fruit of cumin, but also the process applied and the extraction time. The current study showed promising results with regard to the use of spices additives as promoters of growth and health, particularly at the higher incorporation levels in the diet, which are comparable to diets containing probiotics or prebiotics. Cumin can be used as a growth promoter for improving the utilization of food, it can also be used as an antimicrobial agent.

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