

CASE REPORT

Antivirulent-bacterial action of Saudi *Nigella sativa* L Seeds-oil through Corona-pandemic

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Abstract

Background: *Nigella sativa* L named black cumin or black seed, known as “Habbat El Baraka” is grown in Arab desert, and Asia. *Nigella sativa* L oil extract is used to treat infectious diseases caused by virulent bacteria. The aim was to evaluate the effects of Saudi *Nigella sativa* L seeds oil extract against virulent bacteria through Corona pandemic.

Materials and Methods: Preparation of Saudi *Nigella sativa* L seeds oil, and virulent bacteria, then did experience steps. After that were determined turbidity, biomass wet and dry weight.

Results: The results showed the turbidity were higher with all concentrations for Gram-negative than Gram-positive virulent bacteria. The highest turbidity was with *Klebsiella pneumoniae*, *Salmonella typhi*, also *Pseudomonas aeruginosa*. The less turbidity was in *Escherichia coli*. The biomass wet weight was higher with all concentrations for Gram-negative than Gram-positive virulent bacteria. The highest biomass wet weight was in *Klebsiella pneumoniae*, *Salmonella typhi*, also *Pseudomonas aeruginosa*. The less was to *Escherichia coli*. The highest biomass dry weight was on *Klebsiella pneumoniae*, *Salmonella typhi*, also *Pseudomonas aeruginosa*, then *Escherichia coli*. That indicated the more effect on virulent-bacteria were oil on concentration 20%. Considered as the Minimum Inhibitory Concentration (MIC).

Conclusion: The effective Saudi *Nigella sativa* L seeds oil extract concentration was 20%. The extract was more effective in contradiction of Gram-positive bacteria likened to Gram-negative virulent bacteria. Based on these results, Saudi *Nigella sativa* L seeds oil extract could be used as a therapeutic alternative against certain virulent bacterial pathogens. It is up to the “Pharmacy Department” to estimate the most effective dose and concentration of the extract for use in humans.

Keywords: *Nigella sativa* L; Virulent bacteria; Gram-negative bacteria; Gram-positive bacteria

Introduction

Nigella sativa L is an herbaceous plant, known as “Habbat El Baraka”, and its grown in Arab desert. The oil treated infectious diseases caused by virulent-bacteria [1]. The seeds-oil is rich in fixed oils 32-40%, volatile oil 0.4-0.45%, and essential amino acids 8-9 types [2]. Also, alkaloids, steroids, saponins, terpenes, monoterpenes and phenolic [3]. Nigellicine, N-oxide, carvone, thymoquinone, thymol nigellicimine, and nigellicimine are present [4]. The antivirulent-bacteria activities related to fatty acids, and essential oil [5]. Physical cause variation in phytoconstituents percent leads to modification in biological act [6]. The oil treats skin inflammation, and virulent-bacterial in topical infections [7], that for chemical compounds [8-9]. The antivirulent-bacterial activities were to *Escherichia coli*, as well to *Salmonella* spp., [10], *Salmonella typhi* [11], and to numerous antibiotics-resistant bacteria *Staphylococcus aureus* [12-14]. The oil antivirulent-bacteria assist in food store; it was at 2.0% inhibited 24 virulent-bacteria [15]. Also, were on *Escherichia coli*, *Staphylococcus* spp., *Klebsiella pneumoniae* as well on *Salmonella typhi* [16]. Its acted-on *Streptococcus pyogenes*, *Pseudomonas aeruginosa*, as well *Klebsiella pneumoniae*. The greater was against the Gram-positive virulent-bacteria [17]. Thymo-quinone oil act on *Listeria monocytogenes*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Escherichia coli*, and *Salmonella typhi*. It was able to prevent virulent-bacterial biofilm formation [2]. The oils in KSA at 2017 showed maximum antivirulent-bacterial activity against *Escherichia coli* due to compounds. The components were as an alternate medicine and herbal traditional treatment [18]. At 2021, Saudi black oils had antivirulent-bacterial action for *Staphylococcus aureus*, as antibiotics alternate [19]. The aim was using Saudi *Nigella sativa* L seeds-oil as herbal treatment on virulent-bacteria due to prevention secondary infections. That was for its antivirulent-bacteria possessions. So that was to prove the person health prevention during Corona-pandemic.

Materials and Methods

Preparation of Experience Items: Saudi *Nigella sativa* L Seeds-oil was purchased from the “Approved Al-Atara Market Stores”, as use conventional medicine, was much than 98%. The oil was diluted by Ethanol 99.9% to (5, 10, 15 and 20%) [20]. The identified virulence-bacteria were brought from the “Bacterial Laboratory”. They were *Staphylococcus aureus*, *Streptococcus pyogenes*, as well *Streptococcus pneumoniae*, *Listeria monocytogenes*, *Escherichia coli*, also *Klebsiella pneumoniae*, as well *Salmonella typhi*, and *Pseudomonas aeruginosa*. They were cultured on Mueller Hinton Agar (Lab M Ltd., UK), then were grown in Peptone Water (Oxoid, Basingstoke, UK), for suspension matched the 0.5 McFarland to (1.5×10^8 CFU / mL) [21].

Experience Steps: Five ml of virulent-bacterial suspension was added, then one ml of Saudi *Nigella sativa* L seeds-oil diluted. Then were incubated overnight at 37°C [22]. The first test tubes were determined turbidity by “McFarland Standard” [23]. The second test tubes culture was centrifuged to pellets and were weight [24]. The third test tubes cells were centrifuged to pellets. Then the were dried and were weight [24].

Data Analysis: The results were preserved (IBM, Armonk, NY, USA) [25].

Results and Discussions

Turbidity: After overnight incubation at 37°C, the highest turbidity was with *Klebsiella pneumoniae*, *Salmonella typhi*, as well *Pseudomonas aeruginosa*. The less turbidity was in *Escherichia coli*. This indicated the effectiveness on the Gram-positive were more than on the Gram-negative (Table 1). That indicated the more effect on virulent-bacteria were oil on concentration 20%. Considered as the Minimum Inhibitory Concentration (MIC). The oil chemical compounds had antivirulent-bacterial drugs [8-9], to *Escherichia coli*, as well *Salmonella* spp., [10], and *Salmonella typhi* [11]. Also, to multiple antibiotics-resistant bacteria *Staphylococcus aureus* [12-14]. It had on *Escherichia coli*, *Staphylococcus* spp., *Klebsiella pneumoniae* and *Salmonella typhi* [2, 16]. *Pseudomonas aeruginosa* [17]. The oils in KSA at 2017 antivirulent-bacterial activity was against *Escherichia coli* [18]. At 2021, Saudi black oils had acted-on *Staphylococcus aureus*, [19].

Virulent bacteria	Nigella sativa L seeds oil concentration after overnight incubation at 37°C			
	5%	10%	15%	20%
Staphylococcus aureus	0.9±0.1	0.8±0.2	0.7±0.1	0.6±0.2
Streptococcus pyogenes	0.9±0.1	0.8±0.2	0.7±0.1	0.6±0.2
Streptococcus pneumoniae	0.9±0.2	0.8±0.2	0.7±0.1	0.6±0.2
Listeria monocytogenes	0.9±0.1	0.8±0.2	0.7±0.1	0.6±0.2
Escherichia coli	0.9±0.1	0.9±0.1	0.8±0.2	0.7±0.1
Klebsiella pneumoniae	0.9±0.1	0.9±0.1	0.9±0.1	0.8±0.2
Salmonella typhi	0.9±0.1	0.9±0.1	0.9±0.1	0.8±0.2
Pseudomonas aeruginosa	0.9±0.2	0.9±0.2	0.9±0.1	0.8±0.2

Table 1: Turbidity signal of virulent bacteria after overnight incubation at 37°C

Biomass wet weight: After overnight incubation at 37°C, the biomass wet weight was higher for Gram-negative than Gram-positive virulent-bacteria. The highest was in Klebsiella pneumoniae, Salmonella typhi, also Pseudomonas aeruginosa. The Gram-positive were more effected than the Gram-negative (Table 2). That indicated the more effect on virulent-bacteria were oil on concentration 20%. Considered as the Minimum Inhibitory Concentration (MIC). The oil had antivirulent-bacterial drugs [8-9], was to Escherichia coli, as well Salmonella spp., [10], and Salmonella typhi [11], also to multiple antibiotics-resistant bacteria Staphylococcus aureus [12-14]. It had antivirulent-bacterial effects on Escherichia coli, also Staphylococcus spp., Klebsiella pneumoniae and Salmonella typhi [16]. As well, to Streptococcus pyogenes, Pseudomonas aeruginosa, and Klebsiella pneumoniae, the greater against the Gram-positive virulent-bacteria [17]. As well on Listeria monocytogenes, Staphylococcus aureus, Pseudomonas aeruginosa, Escherichia coli, and Salmonella typhi. It was able to prevent virulent-bacterial biofilm arrangement [2]. The oils in KSA at 2017 showed activity against Escherichia coli an alternate medicine and herbal traditional treatment [18]. At 2021, Saudi black cumin oils had antivirulent-bacterial activity on Staphylococcus aureus, alternate to antibiotics for infections care [19].

Virulent bacteria	Nigella sativa L seeds oil concentration after overnight incubation at 37°C			
	5%	10%	15%	20%
Staphylococcus aureus	1.0±0.3	0.7±0.1	0.4±0.2	0.2±0.2
Streptococcus pyogenes	1.0±0.2	0.6±0.2	0.4±0.2	0.2±0.1
Streptococcus pneumoniae	1.0±0.1	0.7±0.2	0.5±0.2	0.2±0.3
Listeria monocytogenes	1.1±0.1	0.7±0.2	0.5±0.1	0.2±0.3
Escherichia coli	1.1±0.1	1.1±0.1	0.8±0.2	0.5±0.3
Klebsiella pneumoniae	1.6±0.2	1.6±0.2	1.6±0.1	1.3±0.2
Salmonella typhi	1.6±0.3	1.5±0.2	1.4±0.1	1.2±0.1
Pseudomonas aeruginosa	1.6±0.4	1.5±0.3	1.5±0.2	1.3±0.1

Table 2: Biomass wet weight by gram of virulent bacteria after overnight incubation at 37°C

Biomass dry weight: After overnight incubation at 37°C, the highest biomass dry weight was on Klebsiella pneumoniae, Salmonella typhi, also Pseudomonas aeruginosa, then Escherichia coli. The effects were less on Gram-negative virulent-bacteria. That showed all concentration provided the most biomass dry weight effect, that was less owing to the oil effect on all virulent-bacteria (Table 3). That indicated the more effect on virulent-bacteria were oil on concentration 20%. Considered as the Minimum Inhibitory Concentration (MIC). The oil had antivirulent-bacterial drugs [8-9], to Escherichia coli, as well Salmonella spp., [10], and Salmonella typhi [11], and Staphylococcus aureus [12-14]. It had antivirulent-bacterial effects on Escherichia coli, Staphylococcus spp., Klebsiella pneumoniae and Salmonella typhi [16-17]. Thymoquinone oil content displayed act on virulent-bacteria [2]. The oils in KSA at 2017 showed maximum antivirulent-bacterial activity against Escherichia coli [18]. At 2021, Saudi black cumin oils had antivirulent-bacterial action on Staphylococcus aureus [19].

Virulent bacteria	Nigella sativa L seeds oil concentration after overnight incubation at 37°C			
	5%	10%	15%	20%
Staphylococcus aureus	0.6±0.1	0.4±0.2	0.2±0.2	0.1±0.2
Streptococcus pyogenes	0.6±0.2	0.4±0.1	0.3±0.1	0.1±0.1
Streptococcus pneumoniae	0.6±0.2	0.5±0.2	0.3±0.2	0.1±0.1
Listeria monocytogenes	0.7±0.1	0.5±0.1	0.3±0.1	0.1±0.1
Escherichia coli	0.7±0.2	0.7±0.1	0.5±0.1	0.3±0.1
Klebsiella pneumoniae	1.0±0.1	1.0±0.1	0.9±0.2	0.8±0.2
Salmonella typhi	0.9±0.2	0.9±0.1	0.9±0.1	0.7±0.2
Pseudomonas aeruginosa	1.0±0.2	0.9±0.2	0.9±0.2	0.8±0.2

Table 3: Biomass dry weight by gram of virulent bacteria after overnight incubation at 37°C

Conclusions

The results were found the effective of Saudi *Nigella sativa L* seeds-oil concentrations were 20%. It continued the effects on the Gram-positive additional than Gram-negative virulent-bacteria through Corona-pandemic.

Recommendations

It was optional the Saudi *Nigella sativa L* seeds-oil usage as a therapeutic alternative for virulent-bacteria. It is up to the “Pharmacy Department” to estimate the quantity and concentration for human using doses.

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