The Beneficial Use of Nigella Sativa Powder in the Healing of Different Portions of Extracted Tooth Socket: In Vivo Study

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ABSTRACT

Back ground: Nigella sativa is so beneficial due to it's content of over a hundred components such as aromatic oils, trace elements, vitamins and enzymes. It contains 58% of essential fatty acids including omega 6 and omega 3. These are necessary for the forming of Prostaglandin E1 which balances and strengthens the immune system giving it the power to prevent infections and allergies and control chronic illnesses. Blackseed oil also contains about 0.5 - 1.5% volatile oils including nigellone and thymochinone which are responsible for its anti-histamine, anti-oxidant, anti-infective and broncho-dilating effect.

Aim of the study: To evaluate the effect of Nigella sativa (powder and oil) on the healing process in different portions of rabbit socket by histomorphometric analysis of bone architecture parameters.

Materials and Methods: The sample of the study consist of twenty rabbits, extract their upper two central incisors under general anesthesia. The left side filled once with Nigella sativa powder and once with Nigella sativa oil material, and the right side left for normal healing as a control group. The two sockets were sutured. The results were studied in intervals period 2 and 6 weeks, histologically and then assayed for histomorphometic analysis for trabecular width, trabecular number and the trabecular separation Marrow space star volume analysis (V*m), Osteoblast, osteocyte, and blood vessels number, in coronal, middle and apical portions of extracted tooth socket. These parameters are derived from microscopic two dimension image measurements analyzer software program.

Results: Histomorphometric analysis for variables of trabeculae width (thickness of trabeculae), trabecular separation and cortical thickness in three studied groups for coronal, middle and apical portions of socket showed significant differences in NgS powder and oil groups in comparison to control. Bone marrow star volume showed a significant variation between NgS powder and NgS oil in comparison to control, also bone marrow star volume showed decrease in it's value with increment in healing period in all groups. Trabecular number and width shows to be higher in NgS powder group in all portions of the socket in comparisum to control and NgS oil groups at 2 and 6 weeks duration. Blood vessels' number shows to be higher in middle portion in NgS powder group at 2 and 6 weeks duration in comparisum to NgS oil and control groups.

Conclusion: Nigella sativa powder suggest to be more effective and enhancer in acceleration of bone formation than oil.

KEYWORDS: Nigella sativa powder, extracted tooth , exodontia , periodontics , healing.

INTRODUCTION

Nigella sativa contains fifteen amino acids; these include eight of the nine essential amino acids required for a healthy diet. *Nigella sativa* oil may also contain parts of the essential oil,[1,2] mostly thymoquinone, by which it acquires an aromatic flavor. However, different brands contain varying chemical compositions.[3] Thymoquinone (TQ) is the predominant bioactive constituent present in black seed oil, it has potential in therapeutic strategy.[4]

MATERIALS & METHODS

The sample of the present study consists of twenty rabbits subjected to extract their upper two central incisors under general anesthesia. The left side filled once with Nigella sativa powder and once with Nigella sativa oil material, and the right side left for normal healing as a control group. The two sockets were sutured. The animals were grouped into: 1st group (10) rabbits, left socket of each rabbit filled by (0.068g) of Nigella sativa powder till 2/3 of socket length which had average length (2.3mm) after removal of blood clot 2nd group (10) rabbits, left socket treated with Nigella sativa oil by using a piece of cotton impregnated in oil then inserted inside the socket till 2/3 of socket length after removal of blood clot and waiting for five minutes then removed it. Each group divided into two subgroups (5 in number) with healing periods; 2 weeks and 6 weeks and the socket examined histologically under light microscope.

Parameters Currently Used To Measure Trabecular Bone Microarchitecture

The microarchitecture descriptors are: trabecular width (Tb.Wid, in microns), trabecular number (or more exactly trabecular density) (Tb.N in per millimetre) and the trabecular separation (Tb.Sp, in microns), marrow space star volume(was calculated as $V^*m=\pi/3 \times \text{mean}(L3)$, where L is the distance from a random point to the point intercepted by the trabecular bones lower V*m values indicate better trabecular bone connectivity). These parameters are derived from microscopic two dimension image measurements analyzer

software program (Vesterby *et al.* 1989). Measurement of trabecular bone microarchitecture by histomorphometric methods based on mathematical morphology and is calculated as the average of the all slices values. [5] Osteoblast, osteocyte, and blood vessles numbers calculated as the average of the all slices values for each period and for each group.

Statistical Analysis

All the data of the sample were subjected to computerized statistical analysis using SPSS version 15 (2006) computer program. The statistical analysis included:

Descriptive Statistics

- Mean
- Standard deviation (SD)

Inferential Statistics

- ANOVA test: for the comparison among the groups.

Least significant difference test LSD test: for variables that show significant differences among the study groups in ANOVA test.

RESULTS

Histomorphmetric analysis of bone architecture parameters in coronal, middle and apical portions of a socket of different studied groups in different periods illustrate the followings:

1. At 2 Weeks Duration

A. Coronal portion

According to the ANOVA (table 1), a highly significant difference was found in TBWID, TBSEP, CORWID and V while a significant difference was found in OBNO and OCNO only. Regarding to the LSD (table 2) for the mean of variables, a highly significant difference in TBWID, CORWID, OBNO and OCNO between NS powder in comparison with the control group and a highly significant difference in TBWID between NS powder and NS oil,The NS oil shows a significant difference in TBWID and CORWID and a highly significant difference in OCNO in comparison with the control group. The control group found to have a highly significant difference in TBSEP compared with NgS oil and powder; and a significant difference in V value in comparison with the NS powder. (Tables 1,2)

B. Middle portion

The ANOVA table (3-6) showed a highly significant difference in (TBWID, TBSEP, CORWID, OBNO and V); and a significant difference in BVNO only. According to the LSD table (3-7), the NgS powder shows a highly significant difference in TBWID, CORWID and OBNO compared to the control group and a highly significant difference in OBNO and CORWID and BVNO in comparison with the NS oil. On the other hand, the NS oil found to have a significant difference in TBWID and CORWID; and a highly significant difference in OBNO compared with the control group while, in comparison with NgS powder, the NgS oil shows a highly significant difference in TBSEP and BVNO. Finally, the control group found to have a highly significant difference in TBSEP and V value compared with the both NgS oil and powder. (Tables 3,4)

C. Apical portion

According to the ANOVA table (3-8), a highly significant difference was found in all the variables except BVNO showed a no significant difference. The LSD table (3-9) shows a significant difference in TBNO and a highly significant difference in TBWID, CORWID, OBNO and OCNO between the NS powder and the control group, While the NgS powder shows a significant difference in TBWID CORWID, OBNO and OCNO when compared with the NS oil. The NS oil found to have a significant difference in CORWID and a highly significant difference in TBNO compared with the control group. Finally, the control group revealed a highly significant difference in TBSEP and V when compared with both the NgS oil and powder. (Tables 5,6)

2. At 6 Weeks

A. Coronal portion

According to the ANOVA table (3-16), a highly significant difference was found in TBNO, TBWID, TBSEP, CORWID and V. The LSD table (3-17) shows that the NgS powder had a highly significant difference in TBNO compared to the control group and a significant difference in TBNO also compared with NS oil, while a highly significant difference was found in TBWID and CORWID in comparison with the control and NgS oil. On the other hand, the NgS oil was found to have a significant difference in TBWID compared to the control group and a highly significant difference in TBSEP and V compared to the NgS powder. Lastly, the control group shows a highly significant difference in TBSEP and V compared to the NgS oil and powder. (Tables 7,8)

B. Middle portion

Regarding the ANOVA table (3-18) shows a significant difference in TBNO and a highly significant difference in the rest variables were found

among the groups. The LSD table (3-19) revealed that the NS powder had a significant difference in TBNO and a highly significant difference in TBWID, CORWID, OBNO and OCNO compared to the control group and NgS oil, and a significant difference was found in BVNO compared to the control group and the NgS oil. The NgS oil shows a significant difference in CORWID compared to the control group and a highly significant difference in OBNO and V compared with the control and NgS powder respectively. Finally, the control group shows a highly significant difference in TBSEP and V values compared to the NgS oil and powder. (Tables 9,10)

C. Apical portion

According to the ANOVA table (3-20), a highly significant difference was found in all variables except in BVNO which was found with no significant difference. The LSD table (3-21) revealed that in the NgS powder there was a highly significant difference in TBNO, OBNO and OCNO in comparison with the control group and NgS oil, while it had a highly significant difference in TBWID and CORWID when compared with the control group and a significant difference in CORWID only when compared with NgS oil. The NgS oil was found to have a significant difference in TBWID and a highly significant difference in CORWID compared to the control group. On the other hand, the control group shows a highly significant difference in TBSEP compared with the NgS oil and powder; and a significant difference in V value compared to the NgS oil and a highly significant difference in the same variable compared with the NgS powder. (Tables 11,12)

DISCUSSION

The effect of Nigella sativa seed extracts on varies body systems was studied *in vitro* and *in vivo*. The pharmacological investigation of the seed extracts reveal abroad spectrum of activities including immuneopotentiation, antihistaminic, antidiabetic, antihypertensive, antinflammatory, antitumor[6] antiparasitic, antibacterial, antifungal and antioxidant.[7] In recent study, the black seed oil extract induced bone healing as manifested by faster bone trabeculae formation and mature bone formation.[8]

Histomorphometric Findings:

The equality of means and variances of all parameters tested for most of micro architecture records between all contrasted study groups include experimental NgS (powder and oil) and control in all different portions (coronal,middle and apical) illustrate a high value in experimental than in control. This result can be explained on a fact that NgS has Thymoquinone (TQ) which is the predominant bioactive constituent present in black seed oil.[9] It has potential in enhancement and recruitment of the progenitor cell to be differentiated to osteoblast cell (bone formative cell) and then trabecular bone formation.[10,11]

Results conclude that there was a significant difference in bone architecture parameters in different intervals time. Increment in trabeculi width, trabeculi number and cortical width is shown in the NgS powder group in comparison to oil and control groups at 2 and 6 weeks duration in all tooth socket portions and it could be attributed to a fact that powder NgS will not lose any of its chemical component proteins, amino acid, aromatic oils, trace elements, vitamins and enzymes, essential fatty acids including omega 6 and omega 3 in comparisum to prepared oil NgS that records low volatile oils content.[12,13]

Blood vessels number presented to be highly significant in middle portion for NgS powder group in comparisum to NgS oil and control groups, it could be explained into:[14,15]

1. NgS itself enhance angiogenesis in area of bone formation but using of NgS in oil form and for 5 mints will not allow for complete absorption by surrounding tissue so it reflects on its effect when it compared with NgS powder.
2. The socket form of the anterior tooth of the rabbit is curved concerned with middle potion and therefore powder of NgS impacted in the middle and shows more effectiveness of its value than the oil one.

CONCLUSION

Nigella Sativa powder is seem to be more effective than oil one in the healing of extracted socket of rabbit as the results illustrate thicker trabecular width and more in number in all socket portions include coronal, middle and apical as well as the number of blood vessels in the middle portion exclusively.

(Table 1) Comparison the coronal portions of (control, NgS oil and NgS powder within 2nd week)

ANOVA

Variables	F	Sig.
TBNO	1.357	.305
TBWID	31.500	.000
TBSEP	45.345	.000
CORWID	9.380	`.006
OBNO	6.796	.016
OCNO	8.000	.010
BVNO	3.800	.064
V	10.555	.004

df = 2

(Table 2) Multiple Comparisons LSD

Variable	Group		Mean Difference	Sig.
TBWID	1	2	-.75	.029
		3	-2.25	.000
	2	3	-1. 50	.001
TBSEP	1	2	9.50	.000
		3	11.00	.000
	2	3	1.50	.262
CORWID	1	2	-2.75	.028
		3	-4.50	.002
	2	3	-1.75	.129
OBNO	1	2	-3.00	.065
		3	-5.25	.005
	2	3	-2.25	.150
OCNO	1	2	-2.00	.007
		3	-2.00	.007
	2	3	.00	1.000
V	1	2	.50	.990
		3	150.50	.003
	2	3	150.00	.003

** The mean difference is significant at the .05 level. (1=coronal control, 2=coronal oil, 3=coronal powder)*

(Table 3) Comparison the middle portions of (control, NgS oil and NgS powder within 2nd week)

Variables	F	Sig.
TBNO	2.000	.191
TBWID	11.382	.003
TBSEP	70.099	.000
CORWID	16.846	.001
OBNO	85.846	.000
OCNO	.661	.540
BVNO	7.824	.011
V	25.381	.000

df = 2

(Table 4) Multiple Comparisons LSD

Variable	Group		Mean Difference	Sig.
TBWID	1	2 3	-2.00 -3.25	.017 .001
	2	3	-1.25	.102
TBSEP	1	2 3	5.50 11.75	.000 .000
	2	3	6.25	.000
CORWID	1	2 3	-1.75 -4.25	.041 .000
	2	3	-2.50	.008
OBNO	1	2 3	-8.50 -20.00	.000 .000
	2	3	-11.50	.000
BVNO	1	2 3	-1.00 2.25	.265 .025
	2	3	3.25	.004
V	1	2 3	127.50 162.50	.000 .000
	2	3	35.00	.179

** The mean difference is significant at the .05 level. (1=Middle control, 2=Middle oil, 3=Middle powder)*

(Table 5) Comparison the apical portions of (control, NgS oil and NgS powder within 2nd week)

ANOVA

Variables	F	Sig.
TBNO	8.591	.008
TBWID	9.632	.006
TBSEP	35.354	.000
CORWID	24.761	.000
OBNO	20.192	.000
OCNO	26.000	.000
BVNO	.000	1.000
V	31.707	.000

df = 2

(Table 6) Multiple Comparisons LSD

Variable	Group		Mean Difference	Sig.
TBNO	1	2	-2.25	.003
		3	-1.50	.024
	2	3	.75	.208
TBWID	1	2	-1.00	.083
		3	-2.25	.002
	2	3	-1.25	.038
TBSEP	1	2	3.50	.009
		3	8.75	.000
	2	3	5.25	.001
CORWID	1	2	-2.75	.019
		3	-6.75	.000
	2	3	-4.00	.002
OBNO	1	2	-1.25	.067
		3	-3.75	.000
	2	3	-2.50	.002
OCNO	1	2	-1.00	.117
		3	-4.00	.000
	2	3	-3.00	.001
V	1	2	125.00	.000
		3	175.00	.000
	2	3	50.00	.055

** The mean difference is significant at the .05 level.*
(1=Apical control, 2=Apical oil, 3=Apical powder)

(Table 7) ANOVA test of coronal portions in (control, NgS oil and NgS powder within 6th week)

Variables	F	Sig.
TBNO	9.176	.007
TBWID	33.876	.000
TBSEP	121.091	.000
CORWID	133.300	.000
OBNO	1.714	.234
OCNO	.339	.721
BVNO	.391	.687
V	59.303	.000

df = 2

(Table 8) Multiple Comparisons LSD

Variable	Group		Mean Difference	Sig.
TBNO	1	2	-.50	.330
		3	-2.00	.003
	2	3	-1.50	.013
TBWID	1	2	-6.25	.043
		3	-21.25	.000
	2	3	-15.00	.000
TBSEP	1	2	6.00	.000
		3	10.50	.000
	2	3	4.50	.000
CORWID	1	2	-3.00	.001
		3	-10.25	.000
	2	3	-7.25	.000
V	1	2	57.50	.001
		3	127.50	.000
	2	3	70.00	.000

** The mean difference is significant at the .05 level. (1=Apical control, 2=Apical oil, 3=Apical powder)*

(Table 9) ANOVA test of middle portions in (control, NgS oil and NgS powder within 6th week)

Variables	F	Sig.
TBNO	5.727	.025
TBWID	51.233	.000
TBSEP	15.429	.001
CORWID	22.389	.000
OBNO	52.043	.000
OCNO	18.048	.001
BVNO	13.364	.002
V	25.038	.000

df = 2

(Table 10) Multiple Comparisons LSD

Variable	Group		Mean Difference	Sig.
TBNO	1	2	.25	.538
		3	-1.00	.031
	2	3	-1.25	.011
TBWID	1	2	-1.75	.177
		3	-11.25	.000
	2	3	-9.50	.000
TBSEP	1	2	3.00	.005
		3	4.50	.000
	2	3	1.50	.102
CORWID	1	2	-2.25	.029
		3	-5.75	.000
	2	3	-3.50	.003
OBNO	1	2	-3.25	.000
		3	-5.75	.000
	2	3	-2.50	.002
OCNO	1	2	-1.75	.094
		3	-5.50	.000
	2	3	-3.75	.003
BVNO	1	2	-.75	.087
		3	-2.00	.001
	2	3	-1.25	.011
V	1	2	40.00	.009
		3	85.00	.000
	2	3	45.00	.005

** The mean difference is significant at the .05 level. (1=Apical control, 2=Apical oil, 3=Apical powder*

(Table 11) Comparison the apical portions of (control, NgS oil and NgS powder within 6th week)

ANOVA

Variables	F	Sig.
TBNO	15.800	.001
TBWID	8.559	.008
TBSEP	10.048	.005
CORWID	29.207	.000
OBNO	38.613	.000
OCNO	33.091	.000
BVNO	.913	.435
V	8.866	.007

df = 2

(Table 12) Multiple Comparisons LSD

Variable	Group		Mean Difference	Sig.
TBNO	1	2	-.75	.135
		3	-2.50	.000
	2	3	-1.75	.004
TBWID	1	2	-2.00	.017
		3	-2.75	.003
	2	3	-.75	.303
TBSEP	1	2	3.50	.005
		3	3.75	.003
	2	3	.25	.795
CORWID	1	2	-5.50	.001
		3	-8.25	.000
	2	3	-2.75	.034
OBNO	1	2	-1.25	.089
		3	-5.50	.000
	2	3	-4.25	.000
OCNO	1	2	-.50	.479
		3	-5.00	.000
	2	3	-4.50	.000
V	1	2	45.00	.038
		3	77.50	.002
	2	3	32.50	.113

** The mean difference is significant at the .05 level.*
(1=Apical control, 2=Apical oil, 3=Apical powder)

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