Impact of *Foeniculum vulgare* on Hemoglobin, Red Blood Cell Indices and White Blood Cell Count: A Laboratory Based Randomized Controlled Study

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Authors’ contributions

This work was carried out in collaboration among all authors. Author AA designed the study, performed the statistical analysis and wrote the protocol. Author RI reviewed and edited the final draft. Author AA supervised. Author QUA managed the analyses of the study. Author FH managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

**Aims:** Anemia is prime health issue of the population in developing region of world mostly due to dietary deficiencies and poor sanitation. Supplementary medicines as well as iron rich foods are utilized to combat this problem. Herbal products also have value in such diets. We evaluated role of *Foeniculum vulgare* incorporated diet on hemoglobin concentration and related indices.

**Study Design:** laboratory centred randomized controlled trial.

**Place and Duration of Study:** Pharmacology Department of University of Karachi, Karachi between June 2018 and September 2018.

**Methodology:** After selecting 30 healthy rabbits of either sex, we placed them in three groups; Control, 2% *Foeniculum vulgare* and 4% *Foeniculum vulgare* group. Control group, 2% and 4% *Foeniculum vulgare* groups were maintained on standard rodent diet, standard rodent diet

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containing 2% and 4% *Foeniculum vulgare* crushed seeds, respectively. Complete blood count of all rabbits was done twice after interval of a month. 

**Results:** In comparison to control group, hemoglobin levels increased in both the study groups, while leucocyte count (WBC) increase was noted only in 4% *Foeniculum vulgare* group. 

**Conclusion:** *Foeniculum vulgare* may have some role in maintenance of hemoglobin levels if not improving it.

**Keywords:** *Foeniculum vulgare*; complete blood count; standard diet; rabbits; seeds.

**1. INTRODUCTION**

Anemia is a common health issue in underdeveloped countries especially affecting women and children. Globally approximately 2 billion human populations suffer from this problem. Anemia is defined as decreased hemoglobin or red blood cell count or both; iron deficiency being the most common cause [1].

About 80% of people around the world use herbal medicine or plant products for many ailments[2]. Since, these remedies possess lesser unwanted effects and are metabolized easily thus, are safer to use [3]. Nearly 2000 ethnic groups all over world utilize herbal remedies according to their traditional therapy knowhow and circumstances [4,5].

Herbal products and extracts are an important source of natural antioxidants such as flavonoids and phenols. These compounds are well known for their therapeutic potential [1]. *Foeniculum vulgare* is one of the many herbs which are used for their therapeutic ability [6]. This perennial plant belonging to Apiaceae family, grows in most regions of globe. It bears small yellow flowers arranged in inflorescence and thin leaves [7]. Its different parts as whole or in extract form are being consumed as remedial agents for illnesses such as lactation [8], leucorrhea, menstrual pain [9], child birth, gastrointestinal disorders [10] and cardiovascular diseases [6]. The major constituents found in this herb are anethole, camphene cymene, estragole, fenchone, myrcene, P-anisaldehyde, α-phellandrene, α-pinene, β-pinene and γ-terpinine [11].

The aim of this study was to assess the impact of *Foeniculum vulgare* seed containing diet on haemoglobin, red blood cell indices and white blood cell count.

**2. MATERIALS AND METHODS**

**2.1 Study Design**

It was a laboratory based randomized controlled study conducted in the Pharmacology Department of University of Karachi. The ethical standards of this study were in accordance with the ARRIVE guidelines and was carried out in accordance with the U.K. Animals (Scientific Procedures) Act, 1986 and associated guidelines, EU Directive 2010/63/EU for animal experiments and the study protocol was approved by Board of Advanced Studies and Research (BASR), University of Karachi, Resol. No. 10(P)14 [12].

**2.2 Plant Material**

*Foeniculum vulgare* seeds were purchased from a local departmental store and identified from the Pharmacognosy Department, Faculty of Pharmacy and Pharmaceutical Sciences University of Karachi, and given the voucher no. FVF-02-15/17.2.2 [13].

**2.3 Animals**

Adult, healthy, albino rabbits (weight 1500-2000gram) of either sex, were taken from the Animal house of Pharmacology department of University of Karachi. Health of all grouped animals was assessed in the acclimatizing duration of a week, particularly looking for any sign of diarrhoea, swelling, lack of activity and hair loss. Rabbits were housed in transparent cages and kept in controlled temperature of 23 ±2°C. Specific diet and water was provided ad libitum for 2 months. Rabbits were placed in 3 groups, Group 1 being the Control group, Group 2 (2% *Foeniculum vulgare*) and Group 3 (4% *Foeniculum vulgare*) were study groups. Control rabbits were given standard diet, while, rabbits belonging to group 2 and 3 were given 2% and 4% *Foeniculum vulgare* seeds containing diet, respectively [14].

**2.4 Sample Collection**

After 30 days interval 2ml blood was collected twice, in K3-EDTA (Ethylene diamine tetra acetic acid) tubes for hematological examination such as red blood cell count (RBC), white blood cells count (WBC), hemoglobin (Hb) and RBC indices.
Complete blood count was conducted on automatic Humacount plus (3 part differential with histogram. Hematology analyser, Model#16400/S) (Human Germany).

2.5 Statistical Analysis

Analysis of results was done by statistical package for social sciences (SPSS) 17.0. Readings were shown as mean ± standard deviation (SD) and were compared by Analysis of variance (ANOVA) followed by post hoc Tukey’s test. P value = 0.05 was considered significant, P value = 0.01 was considered very significant and P value = 0.001 was considered as highly significant.

3. RESULTS

It is evident from resulting values that Foeniculum vulgare incorporated diet increased hemoglobin remarkably in both study groups especially 4% Foeniculum vulgare group. Erythrocyte count increased in both treatment groups whereas, leucocyte count was raised significantly in 4% Foeniculum vulgare group (Table 1).

4. DISCUSSION

Although, medicinal plants have relatively lesser therapeutic actions than synthetic agents, their usage is comparatively more than allopathic medicines in many communities and ethnic groups, due to their lower potential of unwanted effects. An earlier study showed beneficial and positive actions of Foeniculum vulgare hydro-alcoholic extract on red and white blood cell counts. The outcome of the study revealed that extract of Foeniculum vulgare, significantly increased the number of erythrocytes in healthy rats [15]. Study conducted by El-Adawy and colleagues demonstrated similar results with marked increase in erythrocyte count by usage of Foeniculum vulgare fortified diet [16] as our study. Our findings were similar with both of these abovementioned studies with respect to RBC count. Similar results were observed in a study conducted by Tanvir et al who checked hepatoprotective, nephroprotective, anti-inflammatory and anti-ulcerogenic potential of Foeniculum vulgare extracts [17]. Another study by Khushwah and coworkers, showed antianemic effect of this herb where anemia was induced using phenylhydrazine [18]. Owing to the fact that plants possess antioxidant potential, thus, can overcome harmful effect caused by free radicals. Previous multiple studies have proven the deleterious actions of free radicals on erythrocyte cell membrane. Thus, by maintaining the integrity of erythrocytes cell membrane because of their antioxidant action, medicinal plants could be used as therapeutic agent for anemia.

An increase in leucocyte count was observed in our study in animals of both the groups given Foeniculum vulgare incorporated diet and these findings are in accordance to a study by El-Adawy and co-workers [16] who observed the effect on the immune system. The total leucocyte count increase noted in both the treated groups of our study is supported by a study conducted by Mansouri et al [15].

<table>
<thead>
<tr>
<th>Groups</th>
<th>Day 30</th>
<th>Day 60</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control group</td>
<td>8.72±1.75</td>
<td>8.11±1.10</td>
<td></td>
</tr>
<tr>
<td>2% Foeniculum vulgare group (2% FV diet)</td>
<td>9.15±1.93</td>
<td>9.65±0.99</td>
<td>0.012</td>
</tr>
<tr>
<td>4% Foeniculum vulgare group (4% FV diet)</td>
<td>9.25±2.42</td>
<td>11.35±2.48</td>
<td>0.005a</td>
</tr>
</tbody>
</table>

Table 1. Effect of Foeniculum vulgare diet on Hemoglobin

| n=10. Values are mean ± SD, data analysed by one way ANOVA followed by multiple comparison (post hoc Tukey’s test); aP-value is very significant in comparison to control on day 60; bP-value is significant within the group; cP-value is significant among the study groups on day 60 |

<table>
<thead>
<tr>
<th>Groups</th>
<th>Day 30</th>
<th>Day 60</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control group</td>
<td>4.22±1.61</td>
<td>4.10±1.37</td>
<td></td>
</tr>
<tr>
<td>2% Foeniculum vulgare group (2% FV diet)</td>
<td>5.55±1.94</td>
<td>5.94±1.19</td>
<td>0.012a</td>
</tr>
<tr>
<td>4% Foeniculum vulgare group (4% FV diet)</td>
<td>5.64±1.35</td>
<td>6.39±1.12</td>
<td>0.005b</td>
</tr>
</tbody>
</table>

Table 2. Effect of Foeniculum vulgare diet on red blood Cells count

| n=10. Values are mean ± SD, data analysed by one way ANOVA followed by multiple comparison (post hoc Tukey’s test); aP-value is significant in comparison to control on day 60; bP-value is very significant in comparison to control on day 60 |
Table 3. Effect of *Foeniculum vulgare* diet on White Blood Cells count

<table>
<thead>
<tr>
<th>Groups</th>
<th>Day 30</th>
<th>Day 60</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control group</td>
<td>4.09±1.02</td>
<td>4.17±1.29</td>
<td></td>
</tr>
<tr>
<td>2% <em>Foeniculum vulgare</em> group (2% FV diet)</td>
<td>5.14±1.96</td>
<td>4.52±1.44</td>
<td>0.039&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>4% <em>Foeniculum vulgare</em> group (4% FV diet)</td>
<td>7.23±4.10</td>
<td>6.03±1.15</td>
<td>0.001&lt;sup&gt;b&lt;/sup&gt; 0.001&lt;sup&gt;c&lt;/sup&gt; 0.002&lt;sup&gt;d&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

n=10. Values are mean ± SD, data analysed by one way ANOVA followed by multiple comparison (post hoc Tukey’s test); <sup>a</sup>P-value is significant in comparison to control on day 30; <sup>b</sup>P-value is highly significant in comparison to control on day 30; <sup>c</sup>P-value is highly significant among groups on day 30; <sup>d</sup>P-value is very significant among groups on day 60.

Table 4. Effect of *Foeniculum vulgare* diet on Hematocrit

<table>
<thead>
<tr>
<th>GROUPS</th>
<th>Day 30</th>
<th>Day 60</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control group</td>
<td>31.18±2.92</td>
<td>32.03±1.48</td>
<td></td>
</tr>
<tr>
<td>2% <em>Foeniculum vulgare</em> group (2% FV diet)</td>
<td>32.33±1.79</td>
<td>36.86±2.78</td>
<td>0.001&lt;sup&gt;a&lt;/sup&gt; 0.001&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>4% <em>Foeniculum vulgare</em> group (4% FV diet)</td>
<td>33.12±1.54</td>
<td>36.89±2.07</td>
<td>0.002&lt;sup&gt;c&lt;/sup&gt; 0.001&lt;sup&gt;d&lt;/sup&gt; 0.001&lt;sup&gt;e&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

n=10. Values are mean ± SD, data analysed by one way ANOVA followed by multiple comparison (post hoc Tukey’s test); <sup>a</sup>P-value is significant in comparison to control on day 30; <sup>b</sup>P-value is highly significant in comparison to control on day 30; <sup>c</sup>P-value is highly significant in comparison to control on day 60; <sup>d</sup>P-value is highly significant in comparison to control on day 60; <sup>e</sup>P-value is highly significant within group.

Table 5. Effect of *Foeniculum vulgare* diet on RBC indices

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Groups</th>
<th>Day 30</th>
<th>Day 60</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean corpuscular volume</td>
<td>Control</td>
<td>60.88±1.94</td>
<td>60.14±2.89</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2% <em>Foeniculum vulgare</em></td>
<td>62.57±1.69</td>
<td>63.01±2.26</td>
<td>0.05&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>4% <em>Foeniculum vulgare</em></td>
<td>64.20±2.90</td>
<td>60.57±1.47</td>
<td>0.01&lt;sup&gt;b&lt;/sup&gt; 0.05&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>Mean corpuscular hemoglobin</td>
<td>Control</td>
<td>19.88±1.03</td>
<td>19.61±1.95</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2% <em>Foeniculum vulgare</em></td>
<td>18.55±2.15</td>
<td>19.07±2.51</td>
<td></td>
</tr>
<tr>
<td></td>
<td>4% <em>Foeniculum vulgare</em></td>
<td>18.74±1.40</td>
<td>18.09±2.07</td>
<td></td>
</tr>
<tr>
<td>Mean corpuscular Hemoglobin</td>
<td>Control</td>
<td>30.74±2.94</td>
<td>30.21±1.57</td>
<td></td>
</tr>
<tr>
<td>concentration</td>
<td>2% <em>Foeniculum vulgare</em></td>
<td>29.26±2.35</td>
<td>30.87±2.6</td>
<td></td>
</tr>
<tr>
<td></td>
<td>4% <em>Foeniculum vulgare</em></td>
<td>28.79±2.78</td>
<td>30.00±2.54</td>
<td></td>
</tr>
</tbody>
</table>

n=10. Values are mean ± SD, data analysed by one way ANOVA followed by multiple comparison (post hoc Tukey’s test); <sup>a</sup>P-value is significant in comparison to control on day 60; <sup>b</sup>P-value is very significant in comparison to control on day 30; <sup>c</sup>P-value is significant within group.

Elevation in haemoglobin and haematocrit values was revealed in our study. Similarly, increase was noted in some of the indices of RBC in both of our study groups of animals given 2% and 4% *Foeniculum vulgare* fortified diet. These outcomes are similar to a study conducted in Iran where *Foeniculum vulgare* was used as dietary supplement [19] and the study of El Araby and colleagues [20].

Constituents of *Foeniculum vulgare* like coumarins, flavonoids, triglycerides and sterols, have therapeutic potential of being pain relieving, anti-microbial, anti-pyretic and immunomodulatory [21,22]. Antimicrobial effects of *Foeniculum vulgare* ethanolic extract have been noted in multiple studies [23-25]. Anethole a major component found in this herb has demonstrated anti-inflammatory and antioxidant effects and wound healing potential in various experimental models and studies [26-28].

5. CONCLUSION

As it is evident from our study that *Foeniculum vulgare* exert positive effect on haemoglobin and blood cells count and clinical studies could be conducted to verify beneficial effect of this herb.
CONSENT
It is not applicable.

ETHICAL APPROVAL
All authors hereby declare that "Principles of laboratory animal care" (NIH publication No. 85-23, revised 1985) were followed, as well as specific national laws where applicable. All experiments have been examined and approved by the appropriate ethics committee.

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COMPETING INTERESTS
Authors have declared that no competing interests exist.

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16. Mahdavi S, Yeganeh S, Firouzbakhsh F, Janikhalili KH. Effects of Supplementary


