STUDY OF BLOOD COUNT IN THE VALIDITY
EXPERIMENTAL APPENDICITIS IN RABBITS

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SUMMARY: A group of thirty six rabbits was studied, with weight ranging from 2400 to 3100 grams, in order to evaluate the blood test on the validity of experimental acute appendicitis. In order to do that, the animals were distributed among three groups of 12 rabbits each, that corresponded to observation periods of 24 hours (Group A), 48 hours (Group B) and 72 hours (Group C). The presence of macroscopic appendicitis was verified in all the rabbits of the experiment on the three studied periods. The blood test study showed statistic significance when reading the values of haemoglobin, haematocrit and monocytes in Group C; of total and segmented leucocytes, eosinophiles, limphocytes and monocytes in Group B; when the groups faced each other, however, there was no estatistic significance. So, it was concluded that the values of haemoglobin, red blood, white cells and haematocrit are not suitable markers for experimental appendicitis in rabbits.


INTRODUCTION

Acute appendicitis is one of the diseases more frequently approached since its diagnosis misleads the most trained physicians15,16,19,21,29,34,35.

Since 1895 studies have been carried out to produce experimental appendicitis27.

Although mortality rate is low, this disease is a challenge for physicians almost based only on the clinical findings.

In 1996, evaluation of the effects of the ligature of the appendix vermiformis basis of the
rabbit, performed by BAZZANO, led us to decide to investigate blood count in provoked appendicitis in face of a complete occlusion of the appendix vermis of the rabbit⁴.

METHOD

Thirty-six white New Zealand rabbits weighing from 2400 and 3100 grams were used. The animals were divided into three groups of 12 rabbits receiving the denomination of Groups A, B and C.

The animals were put in individual cages seven days previously to the experiment, receiving a commercial ration specially for the specimens and water.

On the day of the experiment the animals received only water. They were intramuscularly anesthetized with acepromazine, xylazine and katamine.

The animal was in decubito dorsalis on the surgical table. After shearing of the auricula sinistra and antisepsis with iodopovidona solution, the extension was performed with exposure through the regio dorsalis of the arteria auricularis sinistra and, blood sample was collected for laboratory analysis.

Through the median laparotomy preceded by shearing of the regio abdominis caudalis, the cecum and appendix vermis were located and ligature of its basis was performed, as proclaimed by BAZZANO in his study⁴.

After the observation period programmed for each group (Group A-24 hours; Group B-48 hours and Group C-72 hours) the animals were anesthetized using the same procedure of the beginning of the experiment. After anesthesia, a new blood sample of the animal was collected. Next, the animals were submitted to an euthanasia and, opening of the cavum abdominis was then performed and macroscopic alterations in the appendix vermis of all animals of the experiment were observed.

Blood samples collected for hematological analysis were forwarded to the laboratory. The compact volume of red blood cells was determined by the microhematocrit method. Hemoglobin was spectrophotometerly measured. The count of red blood and white cells were performed using an automatic cell counter. Differential counts for leukocytes were computed by the method in a glass slide.

For analysis of the results, non-parametric tests were applied considering the nature of the variables studied or variability of the measurements. Wilcoxon Test and Variance Analysis by Kruskal-Wallis rank were performed. In all tests 0,05 or 5% was fixed for level of null rejection, signaling with na asterisk the significant values.

RESULTS

<table>
<thead>
<tr>
<th>TABLE I - Rabbits with appendicitis according to the values of erythrocyte in the observation periods of 24, 48 and 72 hours(x1,000,000)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>24 HOURS</strong></td>
</tr>
<tr>
<td><strong>Before</strong></td>
</tr>
<tr>
<td>4,6</td>
</tr>
<tr>
<td>T&lt;sub&gt;calculated&lt;/sub&gt; = 27,50</td>
</tr>
<tr>
<td>T&lt;sub&gt;critical&lt;/sub&gt; = 11,00</td>
</tr>
<tr>
<td>H&lt;sub&gt;calculated&lt;/sub&gt; = 1,52</td>
</tr>
</tbody>
</table>
**TABLE II - Rabbits with appendicitis according to hemoglobin values in the observation periods of 24, 48 and 72 hours**

<table>
<thead>
<tr>
<th></th>
<th>24 HOURS</th>
<th>48 HOURS</th>
<th>72 HOURS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Before</td>
<td>After</td>
<td>Before</td>
<td>After</td>
</tr>
<tr>
<td>12,7</td>
<td>11,8</td>
<td>12,8</td>
<td>12,1</td>
</tr>
</tbody>
</table>

\[ T_{\text{calculated}} = 20,50 \quad T_{\text{critical}} = 20,00 \quad T_{\text{critical}} = 14,00^* \]

\[ T_{\text{calculated}} = 30,00 \quad T_{\text{critical}} = 14,00 \quad T_{\text{critical}} = 5,99 \]

**TABLE III - Rabbits with appendicitis according to hematocrit in the observation periods 24, 48 and 72 hours**

<table>
<thead>
<tr>
<th></th>
<th>24 HOURS</th>
<th>48 HOURS</th>
<th>72 HOURS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Before</td>
<td>After</td>
<td>Before</td>
<td>After</td>
</tr>
<tr>
<td>40</td>
<td>39</td>
<td>40</td>
<td>37</td>
</tr>
</tbody>
</table>

\[ T_{\text{calculated}} = 31,00 \quad T_{\text{critical}} = 24,50 \quad T_{\text{critical}} = 7,50^* \]

\[ T_{\text{calculated}} = 11,00 \quad T_{\text{critical}} = 14,00 \quad T_{\text{critical}} = 5,99 \]

**TABLE IV - Rabbits with appendicitis according to leukocyte values in the observation periods 24, 48 and 72 hours (x1,000)**

<table>
<thead>
<tr>
<th></th>
<th>24 HOURS</th>
<th>48 HOURS</th>
<th>72 HOURS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Before</td>
<td>After</td>
<td>Before</td>
<td>After</td>
</tr>
<tr>
<td>7,9</td>
<td>6,8</td>
<td>6,7</td>
<td>4,4</td>
</tr>
</tbody>
</table>

\[ T_{\text{calculated}} = 21,00 \quad T_{\text{critical}} = 10,50^* \quad T_{\text{critical}} = 27,00 \]

\[ T_{\text{calculated}} = 14,00 \quad T_{\text{critical}} = 14,00 \quad T_{\text{critical}} = 5,99 \]

**TABLE V - Rabbits with appendicitis according to segmented values in the observation periods of 24, 48 and 72 hours**

<table>
<thead>
<tr>
<th></th>
<th>24 HOURS</th>
<th>48 HOURS</th>
<th>72 HOURS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Before</td>
<td>After</td>
<td>Before</td>
<td>After</td>
</tr>
<tr>
<td>32</td>
<td>29</td>
<td>50</td>
<td>22</td>
</tr>
</tbody>
</table>

\[ T_{\text{calculated}} = 35,00 \quad T_{\text{critical}} = 6,00^* \quad T_{\text{critical}} = 31,50 \]

\[ T_{\text{calculated}} = 14,00 \quad T_{\text{critical}} = 14,00 \quad T_{\text{critical}} = 5,99 \]

**TABLE VI - Rabbits with appendicitis according to eosinophil values in the observation periods of 24, 48 and 72 hours**

<table>
<thead>
<tr>
<th></th>
<th>24 HOURS</th>
<th>48 HOURS</th>
<th>72 HOURS</th>
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</thead>
<tbody>
<tr>
<td>Before</td>
<td>After</td>
<td>Before</td>
<td>After</td>
</tr>
<tr>
<td>22</td>
<td>29</td>
<td>1,8</td>
<td>4,8</td>
</tr>
</tbody>
</table>

\[ T_{\text{calculated}} = 29,00 \quad T_{\text{critical}} = 5,50^* \quad T_{\text{critical}} = 35,50 \]

\[ T_{\text{calculated}} = 14,00 \quad T_{\text{critical}} = 6,00 \quad T_{\text{critical}} = 14,00 \]

\[ T_{\text{calculated}} = 2,01 \quad T_{\text{critical}} = 5,99 \]

**TABLE VII - Rabbits with appendicitis according to lymphocyte values in the observation periods of 24, 48 and 72 hours**

<table>
<thead>
<tr>
<th></th>
<th>24 HOURS</th>
<th>48 HOURS</th>
<th>72 HOURS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Before</td>
<td>After</td>
<td>Before</td>
<td>After</td>
</tr>
<tr>
<td>44</td>
<td>40</td>
<td>46</td>
<td>68</td>
</tr>
</tbody>
</table>

\[ T_{\text{calculated}} = 28,50 \quad T_{\text{critical}} = 6,00^* \quad T_{\text{critical}} = 18,50 \]

\[ T_{\text{calculated}} = 11,00 \quad T_{\text{critical}} = 14,00 \quad T_{\text{critical}} = 5,99 \]

\[ T_{\text{calculated}} = 2,76 \quad T_{\text{critical}} = 5,99 \]
DISCUSSION

Appendicitis is a disease with several clinical presentations and may develop to irreversible disorders up to death if the process is not identified and, its evolution not interrupted by an appendicectomy. Several diseases have signs and symptoms suggesting appendicitis.

In spite of the developments in the medical diagnosis, presently there is no laboratory, radiological tests (or combination of them) or complementary procedure with enough sensibility of effectively diagnose appendicitis in the preoperative period.

In the experimental area, few investigators are concerned in trying to investigate from one model the indicative parameters of the apendicular process.

Anthropoid monkeys have *appendix vermiformis* similar to human being\(^{11,32}\). In these animals which died in bondage, the evidence of spontaneous appendicitis was demonstrated\(^ {30}\).

Thus, dogs and rabbits are used in experimental studies. Due to not secreting mucus as rabbits, monkeys and men do, the *appendix vermiformis* of the dog is considered not suitable for this type of research\(^ {30,31}\).

With the reports of spontaneous appendicitis in 1903, the rabbit is considered the ideal since this animal is easy to be obtained for experimental studies\(^ {23}\).

The use of rabbits in biomedical studies is frequent but still limited for experimental procedures in the surgical area since its postoperative alterations are not known\(^ {8}\).

In spite of the difficulties found for the rabbits, attempts must be done in order to use this animal in surgical procedures due to the following advantages: easy manipulation, docile animals, no need of large spaces for lodging and easy control of diseases\(^ {14,24}\). Rabbits are animals of facile recovering from the effects of anesthetic agents and with an adequate physical shape for many experimental surgical procedures remarkably those needing study of the organs\(^ {24}\).

Hematological parameters were determined for the different periods of the day with no variation in the reading of the blood of white, New Zealand rabbits\(^ {13}\). In order to avoid alterations, the collect of the material was performed in the same period of the day\(^ {13}\).

The values of comprehensible data on the blood constituents (elements) found in the experiment with white, New Zealand rabbits were compared with the results known and catalogued as normal\(^ {10}\).

Confirming the normal parameters in the reading of the erythrocytes of the rabbits\(^ {7,8,18,33}\), by means of ligature of *appendix vermiformis* basis, appendicitis was then provoked on these animals\(^ {4}\) and a slight decrease in the number was observed in the observation periods of 24 hours (group A), 48 hours (group B) and 72 hours (group C).

After the provoked appendicitis in rabbits, the percentage of hematocrit showed a decrease in

| TABLE VIII - Rabbits with appendicitis according to monocytes values in the observation periods of 24, 48 and 72 hours |
|-------------------------------------------|-------------------------------------------|-------------------------------------------|-------------------------------------------|
| Before | After | Before | After | Before | After |
| 24 HOURS | 48 HOURS | 72 HOURS |
| 1,0 | 1,0 | 1,4 | 4,3 | 0,6 | 3,0 |
| \(T_{\text{calculated}} = 16,00\) | \(T_{\text{calculated}} = 3,50^*\) | \(T_{\text{calculated}} = 0,00^*\) |
| \(T_{\text{critical}} = 4,00\) | \(T_{\text{critical}} = 4,00\) | \(T_{\text{critical}} = 4,00\) |
| \(H_{\text{calculated}} = 3,24\) | | \(H_{\text{calculated}} = 5,99\) |
the three groups of the experiment.

Hemoglobin measurements led us to verify the occurrence of a decrease in the Group C concentration.

In the differential counts of the white blood cells, the semented presented a decrease in the count in all the groups with predominance of the Group B, this result followed by the eosinophils.

Regarding lymphocytes, Group A also presented a decrease after provoking appendicitis but in the Groups B and C an increase in the count was observed.

When analyzing monocytes, the counting was increased in all the experiment groups with predominance in the Groups B and C.

Statistical analysis did not show significance in the experiment.

In the investigated literature, no other studies was found regarding the existing alterations in the reading of blood count in rabbits with appendicitis provoked by the ligature of the appendix vermiformis basis.

CONCLUSION

Hemoglobin, red and white blood cells and hematocrit values found in this study are not sufficient to characterize experimental appendicitis in rabbits.

REFERENCES


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RESUMO - A comparação de resultados de exames laboratoriais de coelhos hígidos e coelhos com apendicite provocada, tem como objetivo procurar um marcador para o diagnóstico desta patologia. Foram estudados 36 coelhos com peso corporal variando de 2400 a 3100 gramas, distribuído em três grupos de 12 coelhos, que correspondem aos períodos de observação de 24 horas (Grupo A), 48 horas (Grupo B) e 72 horas (Grupo C). Verificou-se a presença de apendicite macroscópica em todos os coelhos do experimento, nos três grupos estudados. O estudo dos hemogramas mostrou alterações nas leituras dos valores de leucócitos totais, segmentados, eosinófilos, linfócitos e monócitos no Grupo B; da hemoglobina, hematocrito e monócitos no Grupo C; no entanto, quando se confrontam os resultados entre os grupos não há significância estatística. Conclui-se que os valores da hemoglobina, glóbulos vermelhos, glóbulos brancos e hematocrito não são marcadores adequados para apendicite experimental em coelhos.


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