EXPRESSION OF CIRCULATING LEUCOCYTES BEFORE, DURING AND AFTER MYIASIS BY
Dermatobia hominis IN EXPERIMENTALLY INFECTED RATS

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SUMMARY

Expression of circulating white blood cells was investigated in rats (Rattus norvegicus) experimentally infected with larvae of Dermatobia hominis, the human bot fly. Leucocytes were counted prior to infection (control group) as well as at 6, 10, 15, 20 and 28 days post-infection (dpi) and at 7, 15, 30 and 60 days post-larval emergence (dple). Total leucocyte numbers did not differ markedly among the groups. Significant differences were registered when values from control and animals harboring each larval stage of D. hominis were compared; with crescent rank: L1-, L2-, control and L3-infected groups. Leucocyte numbers were significantly higher in the control, 15, 20 or 28 dpi groups than in the 6 dpi animals. Higher counts were observed in control, L2- or L3-infected rats than L1-infected animals. Neutrophils, eosinophils and both large and small lymphocytes were also counted and analyzed. Basophils and monocytes were insufficient in number to permit statistical studies. These results stimulate the continuity of the studies about the host-parasite relationship in the dermatobiosis.

KEYWORD: White blood cells; Myiasis; Dermatobia hominis.

INTRODUCTION

The human bot fly Dermatobia hominis is the most important Neotropical cuterebrid, its larvae producing myiasis in domestic and wild mammals as well as man. The greatest impact of cutaneous myiasis results from economic losses to meat, milk and hide production. Although infections have been systematically combated, the myiasis remains enzootic in South and Central America. Myiasis caused by D. hominis persists for about 40 days in the vertebrate host, causing humoral and cellular changes. However little is known about host humoral and cellular responses.

The present work aimed to monitor the expression of circulating leucocytes before, during and after experimental myiasis by D. hominis and to investigate the relationship between D. hominis larvae and their hosts.

MATERIALS AND METHODS

Five groups of five adult male Rattus norvegicus (Wistar rats) were infected with four newly hatched larvae of D. hominis reared in our laboratory. One sixth group (uninfected animals) was used as control. Smears were made from blood samples taken from the tails of rats from each group: control, at 6, 10, 15, 20 and 28 days post-infection (dpi) and stained with Giemsa. The presence of each larval stage (L1, L2 and L3) associated with host’s blood cell was also analyzed. Blood samples were also collected from rats of each group at 7, 15, 30, 60 days post-larval emergence (dple), and from control group (uninfected animals) as well. The white blood cell differential count (156 fields at 100X magnification) was estimated (at least 200 cells/rat) and the results analyzed by Kruskal-Wallis and Mann Whitney U-test using Winstat software. As a rigged control, all rats (not specific pathogen free) were monitored before infections, including by blood smears.

RESULTS

DAYS POST-INFECTION AND CONTROL

Leucocytes and neutrophils: The total number of leucocytes (Table 1) was not significantly different when compared by Kruskal-Wallis test. Significant differences were observed when larval stages (L1 = 6 dpi, L2 = 10 + 15 dpi) and L3 = 20 + 28 dpi) and control animals were compared, with progressively more leucocytes seen in rats infected with L1, L2, control and finally L3. By U-test, leucocytes counts were significantly higher in the control, 15, 20 or 28 dpi groups than in the 6 dpi one. With respect to larval stages, higher counts were seen in control, L2- or L3-infected groups than in the L1-infected group.

Neutrophils were significant, with progressive number in rats at 6, 10, control, 15, 20 and 28 dpi as well as in those infected with L1, L2, and L3. Between groups, neutrophils were significantly more numerous in the control vs 6 dpi and in 28 dpi vs control. With regard
Leucocytes and neutrophils: No significant differences were found for total leucocytes. Neutrophil numbers increased significantly in the following order of progression: 6 dpi, 30 dpi, 10 dpi, 60 dpi, control, 15 dpi, 60 dpi; and 28 dpi vs 6 dpi and 28 dpi vs 15 dpi. Neutrophil values were significant for 15 and 20 dpi vs 30 dpi; 28 dpi vs 7, 15, 30 and 60 dpi; and 7, 15 and 60 dpi vs 6 dpi.

Eosinophil numbers were progressively higher in groups 28 dpi, 7 dpi, 6 dpi, control, 15 dpi, 60 dpi, 30 dpi, 15 dpi and 60 dpi. Significant differences (between groups) were seen for 15, 30 and 60 dpi vs 28 dpi.

Total numbers of lymphocytes (small plus large) showed marked variation, increasing progressively as follows: 28 dpi, 20 dpi, 15 dpi, 15 dpi, 60 dpi, 7 dpi, 10 dpi, control, 30 dpi and 6 dpi. When two values were compared significant differences were seen for 6 dpi vs 7 dpi and 15 dpi as well as 7, 30 and 60 dpi vs 28 dpi.

Large lymphocytes increased significantly in the following order of progression: 28 dpi, 7 dpi, control, 15 dpi, 20 dpi, 15 dpi, 10 dpi, 60 dpi and 6 dpi. Significant differences were found between the numbers of large lymphocytes for 6 dpi vs 7 and 15 dpi; pro-10 dpi vs 7 dpi; pro-60 dpi vs 15, 20 and 28 dpi.

Small lymphocyte numbers increased significantly in the following order: 60 dpi, 6 dpi, 20 dpi, 28 dpi, 15 dpi, 15 dpi, 30 dpi, 10 dpi, control and 7 dpi. In the comparisons between groups, significant variations of small lymphocyte occurred for 10 dpi vs 60 dpi and 15 dpi vs 28 dpi; and pro-7 dpi vs 6, 20 and 28 dpi.

DAYS POST-LARVAL EMERGENCE AND CONTROL

Leucocytes and neutrophils: No significant differences were found between the total numbers of leucocytes. Significant differences were only seen for neutrophils (U-test): in favour of 7 and 15 vs 30 dpi.

Eosinophils and lymphocytes: Eosinophils were only significant by U-test: pro-15 vs 60 dpi.

Lymphocytes are not significant. However, significant differences were seen for large lymphocytes in the following order: 7, control, 15, 30, 60 dpi and 60 dpi vs control. In comparative analyses of the dpi periods, significant differences were seen for 15, 30 or 60 dpi vs 7 and 60 vs 15 dpi.

The numbers of small lymphocytes were expressive in the following order of progression: 60, 15, 30 dpi, control and 7 dpi. Such cells lymphocytes showed significance in number only for control vs 60 dpi and 30 vs 60 dpi. With regard to dpi, significant differences were seen for 7 vs 15 or 60 dpi and 30 vs 60 dpi.
The present study differs from those observed when the larval stage parasitism are considered, particularly in favour of L_,. These data concur with observations made in cattle infected by *D. hominis*. The leucocyte number here reported is similar to that noted in cattle infected with *D. hominis* but much lower than that cited for *Tamias striatus* naturally infected by *Cuterebra emasculator*. With respect to the specific types of white blood cells the low numbers of neutrophils at the start of infection, as seen in the present study, may be explained by the fact that such cells are the first to arrive at the site of inflammation*. Neutrophilia in the circulation at 28 dpi and during scar formation (7 dpe) was much lower than that seen in the skin during inflammation*. Expression of eosinophilia is more commonly associated with allergic reactions and helminthic parasite infections*. Eosinophils are involved in wound healing and repair, in fibrosis, *e.g.* scarring*. The constant levels of eosinophilia in the blood during myiasis by *D. hominis* seen during the present study differ from those observed in the skin of several human bot fly hosts in previous studies, including rabbit*, cattle*,* and rat*. After the L_ larva dropped from the host to pupate, the number of circulating eosinophils was greater than during infection, although not significantly compared with the control group. Otherwise eosinophilia has recently been observed in sheep infected with the nasal bot fly *Oestrus ovis*. In the present study lymphocytes were similar to those observed in cattle infected with *D. hominis*. Nevertheless, lymphocytosis has been found in bot fly-infected rat skin*. Lymphocytopenia observed in cattle with dermal myiasis by *Hypoderma lineatum* were described*. If compared, the large lymphocyte (lymphoblasts) population remained constant during parasitism by *D. hominis*, but such cells increasing significantly after skin scratching. Nevertheless, small lymphocytes (probably activated cells) fell during myiasis and increased just after the L_ left the host (at 7 dpe). The prevalence of basophils and monocytes in normal rats* were also similar to those found in the present study (insufficient to statistical analysis). In cattle infected with *D. hominis* larvae basophils and monocytes were not expressive*. Basophilia occurs in ectoparasitoses such as sheep scab* and tick*. Knowledge of the variety and numbers of the white cells in host blood during myiasis, through studies such as the one reported here, are essential to focus new research approaches on the relationship between the human bot fly and its hosts.

**REFERENCES**


**ACKNOWLEDGEMENTS**

We thank Bruce Alexander for reviewing this manuscript. This work was partially supported by CNPq and FAPEMIG.

**RESUMO**

Expressão de leucócitos na circulação sanguínea antes, durante e após miíase por *Dermatobia hominis* em ratos experimentalmente infectados

A expressão de leucócitos sanguínea foi investigada em ratos (*Rattus norvegicus*) experimentalmente infectados com larvas de *Dermatobia hominis*. As células foram contadas antes, durante, aos 6, 10, 15, 20 e 28 dias pós-infestaçäo (dpi), e aos 7, 15, 30 e 60 dias pós-emergência das larvas dos hospedeiros. O total de leucócitos não apresentou marcante diferença entre todos os grupos de animais. Todavia, diferenças significativas foram observadas quanto ao parasitismo pelos estádios larvares, com nível crescente: L_, L_ e L_. Na comparação entre grupos: o número de leucócitos foi significativo pró-controle, -15, -20 ou -28 dpi do que aos 6 dpi; e pró-controle, -L_ ou -L_, do que para L_. Neutrófilos, eosinófilos e limfócitos (pequenos e grandes) foram também analisados. Em contraste, o número insuficiente de basófilos e monócitos não permitiram estudos estatísticos. Estes resultados estimulam a continuação dos estudos sobre a relação parasito-hospedeiro nas dermatobioses.


Received: 22 September 2006
Accepted: 16 May 2007