Serial Measurements of Hematologic Counts during the Active Phase of Human Granulocytic Ehrlichiosis

Johan S. Bakken,1 Maria E. Aguero-Rosenfeld,4 Robert L. Tilden,2 Gary P. Wormser,3 Harold W. Horowitz,3 John T. Raffalli,3 Mehdi Baluch,4 Debbie Riddell,3 Jennifer J. Walls,6 and J. Stephen Dumler6

1Section of Infectious Diseases, 2Department of Education and Research, and 3Department of Laboratory Medicine, Saint Mary’s Hospital-Duluth Health System, Duluth, Minnesota; 4Department of Pathology, and Division of Infectious Diseases, 5Department of Medicine, New York Medical College, Westchester Medical Center, Valhalla, New York; and 6Department of Pathology, The Johns Hopkins Medical Institutions, Baltimore

To describe the changes that occur in blood count parameters during the natural course of human granulocytic ehrlichiosis, we designed a retrospective cross-sectional case study of 144 patients with human granulocytic ehrlichiosis and matched controls who had a different acute febrile illness. Patients from New York State and the upper Midwest were evaluated from June 1990 through December 1998. Routine complete blood counts and manual differential leukocyte counts of peripheral blood were performed on blood samples that were collected during the active illness, and values were recorded until the day of treatment with an active antibiotic drug. Thrombocytopenia was observed more frequently than was leukopenia, and the risk of having ehrlichiosis varied inversely with the granulocyte count and the platelet count. Patients with ehrlichiosis displayed relative and absolute lymphopenia and had a significant increase in band neutrophil counts during the first week of illness. Knowledge of characteristic complete blood count patterns that occur during active ehrlichiosis may help clinicians to identify patients who should be evaluated specifically for ehrlichiosis and who should receive empiric antibiotic treatment with doxycycline.

Human granulocytic ehrlichiosis (HGE) is an acute, nonspecific febrile illness that was first described in 1994 among patients who were living in the upper Midwest of the United States [1]. There is increasing evidence of a tick transmission cycle, because patients acquire their infection in areas where Ixodes persulcatus—complex ticks, the presumed tick vectors, are commonly encountered [2–7].

The clinical signs and symptoms of HGE are nonspecific, and although abnormal laboratory values have been reported frequently in the complete blood count (CBC), these findings are similarly nonspecific [8–10]. Acutely infected patients manifest variable reductions in the total WBC and platelet (Plt) counts, and most patients display relative granulocytosis and a left shift [8, 9]. However, patients with documented active HGE may also present with normal WBC or Plt counts [8, 9].

Hossain et al. [11] recently described the serial CBC determinations in a patient with active HGE. However, to our knowledge, little has been published about the sequential changes that occur in CBCs during the natural course of illness. The purpose of this investigation was to extend laboratory monitoring to a larger group of patients with documented HGE infection who were from...
the upper Midwest and from New York State. Our goals were as follows: (a) to determine the proportions of patients who developed anemia, leukopenia, thrombocytopenia, or a combination thereof during the course of illness; (b) to understand the dynamic patterns of change that occur in the absolute concentrations and relative distributions of segmented and band neutrophils, lymphocytes, monocytes, eosinophils, basophils, and Plts during the course of active infection; (c) to assess CBC patterns that might help to identify patients who are possibly infected with the HGE agent who should be considered for further diagnostic testing and empiric doxycycline therapy; and (d) to compare and contrast laboratory findings for patients with HGE who were from the upper Midwest with those for patients with HGE who were from lower New York State.

MATERIALS AND METHODS

**Patient selection.** Medical records of patients who had HGE diagnosed from June 1990 through December 1998 in the upper Midwest (SMDC Health System, Duluth, MN) and in New York (Westchester Medical Center, Valhalla, NY) were reviewed. To be included in the study, all patients had to have had at least 1 CBC determination before they started receiving specific antibiotic treatment. Confirmatory laboratory tests for HGE included 1 or more of the following: light microscopic examination of peripheral blood smears for presence of morulae in granulocytes [8]; indirect immunofluorescent antibody (IFA) tests of paired serum samples, obtained at intervals of at least 2 weeks, for detection of antibodies to *Ehrlichia equi* or the etiologic agent of HGE (hereafter known as “HGE agent”) [8]; identification, by use of PCR, of DNA sequences of the HGE agent in acute-phase blood samples [12, 13]; or isolation of the HGE agent from blood samples in HL-60 human promyelocytic cell culture [14].

Patients were categorized as having probable or confirmed HGE according to previously established case definitions [8]. Probable cases of HGE involved patients who had a history of tick exposure, signs and symptoms of a febrile illness that was compatible with HGE, and (a) presence of morulae in peripheral blood neutrophils, or (b) detectable anti-HGE agent or *E. equi* antibodies in a single serum sample. Confirmed cases of HGE required the presence of a compatible clinical illness and a positive result of PCR analysis of acute-phase blood samples, IFA seroconversion, or in vitro cultivation of the HGE agent from acute-phase blood samples [8]. For the purposes of this article, “patients with HGE” are both those with probable cases and those with confirmed cases.

All patients who were included in the investigation had been either interviewed or examined by one of us (J.S.B., H.W.H., J.T.R., or G.P.W.). The specific information that we recorded included the date of onset of illness, the duration of illness before the diagnosis of HGE was established, and the day of illness on which specific antibiotic treatment was initiated. Days of illness were counted in ascending order, starting with the day of onset of symptoms (day 1). Laboratory test results, when available, were tabulated for each day of illness until (and including) the day of initiation of doxycycline or rifampin therapy or until the patient was lost to follow-up.

**Control patients.** Patients who presented to the SMDC Health System for evaluation of a nonspecific febrile illness that was not caused by HGE were matched retrospectively with 111 study patients from the upper Midwest to determine if the initial CBC could be used to distinguish HGE from other causes of fever. All control patients presented with fever and a history of recent tick exposure or bite, and all had undergone a non-diagnostic physical examination. Each control patient was matched to a study patient according to sex, age (± 5 years), and year of evaluation. HGE was ruled out as the cause of fever if the patient had negative results on blood smear microscopy and absence of antibodies to the HGE agent in acute- and convalescent-phase serum samples. Many control patients also had PCR and blood culture results that were negative for the HGE agent. Most control patients had a self-limited infectious illness of presumed viral etiology.

**Routine laboratory tests.** Acute-phase EDTA blood samples were analyzed by use of a routine Coulter counter. Hemoglobin concentrations, total WBC counts, and Plt counts were recorded. A manual differential count (200 leukocytes counted) was also performed on Wright-stained peripheral blood smears. The relative proportions (expressed as percentages) and absolute concentrations of segmented and band neutrophils, lymphocytes, monocytes, eosinophils, and basophils \((\times 10^9 \text{ cells/L})\) were noted for each patient and for each of the days of illness for which data were available. The relative proportions for leukocytes were included because the differential WBC count expressed by the percentage distribution has been the preferred and traditional reporting system from clinical laboratories to clinicians. Values were compiled for each variable for each day of illness and were expressed as mean values (concentrations or percentage of values) ± SD.

The CBC normal ranges used by the Hematology Laboratory at St. Mary’s Medical Center (Duluth, MN) were used as comparison references for the calculated mean values. Normal ranges for CBC parameter concentrations and relative proportions (as applicable) were as follows: total WBC, 4.0–10.2 \(\times 10^9\) cells/L; segmented neutrophils, 3.2–7.4 \(\times 10^9\) cells/L and 40%–74%; band neutrophils, 0.0–0.6 \(\times 10^9\) cells/L and 0%–6%; lymphocytes, 0.9–3.6 \(\times 10^9\) cells/L and 10%–41%; monocytes, 0.3–1.0 \(\times 10^9\) cells/L and 4%–15%; eosinophils, 0.0–0.6 \(\times 10^9\) cells/L and 0%–8%; basophils, 0.0–0.3 \(\times 10^7\)
cells/L and 0%–3%; Plt, platelets/L; hemoglobin (men), 130–170 g/L; and hemoglobin (women), 117–140 g/L.

Wright-stained acute-phase peripheral blood smears oruffy coat smears were examined by means of light microscopy for detection of morulae, as described elsewhere [8]. A total of 800 granulocytes (or a total of 1000 granulocytes at Westchester Medical Center) were examined before a patient was considered to have a negative blood smear test result.

**Serologic tests.** Serologic testing was performed by use of the IFA method, as described elsewhere [1, 8]. *E. equi* (MRK strain)—infected horse neutrophils, *E. equi* (MRK strain)—infected HL-60 human promyelocyte cells, or the HGE agent (Webster or NY-13 strain)—infected HL-60 human promyelocyte cells were used as marker antigens. Antibody titer values were expressed as the reciprocal of the representative serum dilution, and antibody titers of $\geq 80$ were considered to be a positive result [8, 15, 16].

**Nucleic acid amplification.** Nucleic acids were extracted from acute-phase EDTA blood samples. Either the *Ehrlichia phagocytophila* genogroup-specific primer set ge9f/ge10r or primers for the epank1 gene were used for amplification, as described elsewhere [12, 13].

**Culture of the HGE agent.** EDTA blood samples that were collected from some infected patients during the acute phase of illness were inoculated into HL-60 cells and propagated as described elsewhere [14].

**Definitions.** “Leukopenia” and “leukocytosis” refer to WBC counts of $<4 \times 10^3$ cells/L and $>10.2 \times 10^3$ cells/L, respectively, whereas “thrombocytopenia” and “thrombocytosis” refer to Plt counts of $<150 \times 10^3$ cells/L and $>400 \times 10^3$ cells/L, respectively. The sum of segmented and band neutrophils comprises the total granulocyte level. “Left shift” refers to a band neutrophil concentration or relative proportion above the reference ranges (0.60–10 $\times 10^3$ cells/L and 6%, respectively). “Lymphocytosis” or “lymphopenia” refers to lymphocyte absolute concentrations or relative values above or below the reference range, respectively. “Observation period” refers to the period between day 2 and day 14 of illness.

**Statistical analysis.** All collected data were analyzed by use of SPSS, version 7.5 (SPSS). The Wilcoxon rank sum test, uncorrected $\chi^2$ analysis, $\chi^2$ analysis with Fisher’s exact test correction, Student’s $t$ test, or the Mann-Whitney test was employed to compare independent variables. $P < .05$ was considered to be statistically significant.

**RESULTS**

**Demographic analysis.** A total of 144 patients (96 men [67%] and 48 women [33%]) were included in the investigation. The mean patient age was 56.5 years (SD, 17.9 years). There were no significant differences in mean ages between men and women (56.7 years vs. 56.0 years; $P = .829$). A total of 113 (78.5%) of the patients lived in the upper Midwest, and their mean age was slightly higher than that of the patients who lived in New York (58.0 years vs. 51.1 years; $P = .056$). There was no significant difference in the distribution of men and women between the upper Midwest and New York cohorts (35 [30.9%] of 113 vs. 13 [41.9%] of 31; $P = .285$). The signs and symptoms of HGE had lasted for an average of 5.8 days before doxycycline (135 patients) or rifampin (1 patient) therapy was initiated. Eight patients never received effective antibiotic therapy because the correct diagnosis was not recognized until after the natural illness had abated. The course of untreated HGE lasted for 6–60 days, but most patients felt well and back to normal health by the fourth week after onset of illness.

**Diagnosis confirmation.** The HGE agent was cultured more often in blood samples obtained from infected patients who were from New York State (18 of 19) than those obtained from patients who were from the upper Midwest (11 of 18; $P = .037$). There were no significant differences in the frequencies of positive results of other tests that were used to confirm HGE in patients at each study site (table 1). Morulae were detected in granulocytes from 86 (61.0%) of 141 patients, and 65 (71.4%) of 91 patients had positive PCR results. A total of 119 (97.5%) of 122 patients had seroconversion, whereas IFA titers to the HGE agent that were $\geq 80$ remained stationary for 19 patients. Three patients failed to have seroconversion (2 patients had positive blood culture results, whereas, for the third patient, the diagnosis was confirmed by detection of morulae in neutrophils and a positive PCR result).

**CBC variations.** Laboratory testing was performed during the acute illness for as long as 7 months, but blood samples were collected infrequently after day 14; 5.3% of total samples were collected after day 14. The following report is therefore limited to analysis of data that were collected during the first 2 weeks of illness. Only 1 of 144 patients was evaluated on the 2 weeks of illness.

**Table 1. Results of diagnostic laboratory testing for human granulocytic ehrlichiosis (HGE) and outcomes of tests of 144 patients with HGE from the upper Midwest and from lower New York State.**

<table>
<thead>
<tr>
<th>Laboratory test</th>
<th>Upper Midwest</th>
<th>New York</th>
</tr>
</thead>
<tbody>
<tr>
<td>Morulae in neutrophils</td>
<td>69/113 (61.1%)</td>
<td>17/28 (60.7%)</td>
</tr>
<tr>
<td>Positive PCR result</td>
<td>43/62 (69.4%)</td>
<td>22/29 (75.9%)</td>
</tr>
<tr>
<td>Positive blood culture result</td>
<td>11/18 (61.1%)</td>
<td>18/19 (94.7%)</td>
</tr>
<tr>
<td>Anti-HGE agent IFA titers $\geq 80$</td>
<td>19/19 (100.0%)</td>
<td>2/3 (66.7%)</td>
</tr>
<tr>
<td>$\geq 4$ times change in the anti-HGE agent IFA titers$^a$</td>
<td>91/93 (97.8%)</td>
<td>28/29 (96.6%)</td>
</tr>
</tbody>
</table>

**NOTE.** Data are n/N (%) of patients. IFA, indirect immunofluorescent antibody.

$^a$ Only a single serum sample was available.

$^b$ For acute- and convalescent-phase samples.
Figure 1. Serial variations in mean concentrations of total WBC ($\times 10^9$ cells/L), hemoglobin (g/L), and platelets ($\times 10^9$ cells/L) in peripheral blood samples obtained from 144 patients with active human granulocytic ehrlichiosis during the first 2 weeks of illness before initiation of antibiotic therapy. The test reference ranges are indicated (shaded areas).

Figure 2. Percentage of patients per day who had leukopenia (WBC count, $<4.0 \times 10^9$ cells/L; gray shaded bars) or thrombocytopenia (platelet count, $<150 \times 10^9$ cells/L; black bars) during 2 weeks of active human granulocytic ehrlichiosis and before initiation doxycycline or rifampin therapy. n, Number of patients.

first day of illness. Comparison of mean CBC values for the New York and upper Midwest cohorts failed to show any significant differences, and the analyses reflect the results of pooled data that were collected from patients at both study sites. The normal reference ranges for each CBC parameter are listed in the Materials and Methods section.

A median of 3 separate CBC determinations (range, 1–8) was obtained from each patient during the first 14 days of illness. Both WBC counts and Plt counts were normal for 27 patients (18.8%) on multiple determinations. In contrast, 71 patients (49.3%) displayed both WBC counts and Plt counts that were below the normal range on 1 or more occasions (McNemar $\chi^2 = 26.63; P < .0001$). Forty-one patients (28.5%) had low Plt but normal WBC counts, whereas 5 patients (3.5%) had normal Plt but low WBC counts on 1 or more occasions.

**Total WBC.** The mean WBC concentration decreased from $5.2 \times 10^9$ cells/L on day 2 to a nadir of $3.7 \times 10^9$ cells/L on day 5 ($P = .0003$; figure 1). Thereafter, the mean WBC values slowly increased, and values remained in the normal range from day 6 through day 14. Seventy-six (52.8%) of the 144 patients had WBC values of $<4.0 \times 10^9$ cells/L on 1 or more days during the observation period. Five patients had leukopenia on 3 occasions. Leukopenia was observed most frequently during the first week of illness (figure 2). There was a statistically significant increase in the mean WBC concentrations from day 2 ($5.21 \times 10^9$ cells/L) through day 10 ($7.40 \times 10^9$ cells/L; $P = .017$).

**Hemoglobin.** Mean hemoglobin values decreased from 146 g/L on day 2 to 128 g/L on day 13 ($P = .035$; figure 1). Separate analysis of hemoglobin values for men and women revealed gradually decreasing values for both sexes during the observation period (data not shown). These changes were statistically significant for women ($P = .023$) but not for men ($P = .055$).

**Plt.** The mean Plt concentration was $133 \times 10^9$ cells/L on day 2; it then gradually decreased to a nadir of $81 \times 10^9$ cells/L on day 6 ($P < .0003$). The mean Plt concentration stayed below the normal range until day 9, and thereafter it remained in the normal range for the rest of the observation period (figure 1). By day 11, the Plt concentration had increased to $241 \times 10^9$ cells/L ($P < .00001$). A total of 112 (77.8%) of the 144 patients had thrombocytopenia on 1 or more days during the 14 days of illness. The majority of patients (59.6%–94.1%) had thrombocytopenia on any given day from day 2 through day 9 (figure 2). As late as day 20 or later, Plt counts of $<100 \times 10^9$ cells/L were noted for 2 separate patients.

**Granulocytes.** Although the mean absolute granulocyte
Figure 3. Serial variations in the absolute concentrations of segmented and band neutrophils, and lymphocytes ($\times 10^9$ cells/L) in peripheral blood samples obtained from 144 patients with active human granulocytic ehrlichiosis during the first 2 weeks of illness before initiation of antibiotic therapy. The test reference ranges are indicated (shaded areas).

Figure 4. Serial variations in the relative distributions of segmented and band neutrophils, and lymphocytes (%) in peripheral blood samples obtained from 144 patients with active human granulocytic ehrlichiosis during the first 2 weeks of illness before initiation of antibiotic therapy. The test reference ranges are indicated (shaded areas).

count was within the normal range on day 2, the mean percentage granulocyte count was elevated to 85.1%. This elevation was predominantly the effect of a marked left shift with an increase in the proportion of band neutrophils (figure 3). By day 6, the percentage of granulocytes had decreased to a nadir of 62.9% ($P = .002$). During the same period, the absolute granulocyte concentration decreased from $4.40 \times 10^9$ cells/L to $2.71 \times 10^9$ cells/L ($P = .0014$). Thereafter, the total granulocyte concentrations and relative percentage values fluctuated in the lower half of their respective reference ranges. The relative proportion of patients who had elevated mean total granulocyte differential counts decreased from 75% on day 2 to 7.7% on day 10 (data not shown). There was a significant decrease in the mean relative granulocyte counts between day 2 (85.1%) and day 11 (64.0%; $P = .016$). In contrast, the mean absolute granulocyte concentrations increased from $4.40 \times 10^9$ cells/L on day 2 to $5.34 \times 10^9$ cells/L on day 10, but the change was not statistically significant.

**Segmented neutrophils.** The mean relative proportion of segmented neutrophils remained within the reference range on each day of observation (figure 4). In contrast, the mean absolute concentrations were in the lower normal range for most of the days and below normal on days 4–6 (figure 3). The
proportion of patients who had segmented neutrophil percentage counts below the normal range varied between 6.5% and 34.0% on any given day, and no apparent trends were noted (data not shown).

**Bands.** Both the mean percentage counts and the absolute concentrations of band neutrophils remained elevated above the reference range during the entire observation period, with the exception of a few days during the second week (figures 3 and 4). The mean band percent count decreased from 16.7% on day 2 to 5.6% \( (P = .001) \) on day 11, and the corresponding absolute band concentration also decreased significantly \( (0.89 \times 10^9\, \text{cells/L} \) to \( 0.39 \times 10^9\, \text{cells/L}; \, P = .010 \) \) during the same time. The proportion of patients with elevated relative band counts peaked on day 3 (36 [94.7%] of 38), and the rate thereafter gradually decreased to 20% of patients tested on day 11 (figure 3). A total of 109 (75.7%) of the 144 patients had elevated relative band counts on 1 or more days during days 2–14.

**Lymphocytes.** The mean lymphocyte percentage counts remained within the reference range for each day during the 14 days of illness (figure 4). Only 50 (34.7%) of the 144 patients had relative lymphocyte counts below the normal range on any day from day 2 through day 14 of illness. In contrast, the majority of patients had absolute lymphopenia during the first 5 days (93.4% on day 3 and 73.0% on day 5). The mean absolute lymphocyte concentrations stayed below the reference range from day 2 through day 5 (figure 3). There was a significant increase in the mean lymphocyte concentrations, from \( 0.54 \times 10^9\, \text{cells/L} \) (10.7%) on day 2 to \( 1.36 \times 10^9\, \text{cells/L} \) (30.8%) on day 6 \( (P < .0001) \), and mean values continued to increase during the second week of illness. Although individual lymphocytosis was observed in some cases, the mean absolute lymphocyte count never exceeded the upper reference range limit for any given day. Atypical reactive lymphocytes (>5% of the total lymphocyte count) were noted during the second week of illness in 12 (66.7%) of a subset of 18 patients from the New York cohort.

**Monocytes.** Neither the relative distribution nor the absolute concentrations of monocytes changed significantly during the period of observation. The relative and absolute values remained below or at the lower end of their reference ranges (data not shown). However, these changes were not statistically significant.

**Basophils.** The absolute concentrations and the relative distribution of basophils remained at the lower end of their respective reference ranges for the duration of the observation period, and the complete absence of basophils was noted for several days during the second week of observation (data not shown).

**Evaluation of risk of HGE determined by the initial CBC determination.** The values of the initial CBCs that were collected from 111 patients with HGE from the upper Midwest were compared with those of matched control patients to determine whether abnormal values would be useful to predict the risk of HGE (table 2). Although the mean total WBC concentration was within the normal range for both patient cohorts, it was significantly higher in the control patients than in the patients with HGE \( (P < .00001) \). The initial mean Plt count was normal \( (213 \times 10^9\, \text{cells/L}) \) for the control cohort, but it was moderately reduced for the patients with HGE \( (124 \times 10^9\, \text{cells/L}; \, P < .00001) \).

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value for Control patients</th>
<th>Value for Patients with HGE</th>
<th>( P )</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total WBC count, ( \times 10^9, \text{cells/L} )</td>
<td>7.2 ± 3.8</td>
<td>5.0 ± 2.4</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Segmented neutrophils, ( \times 10^9, \text{cells/L} )</td>
<td>4.8 ± 3.3</td>
<td>3.1 ± 2.0</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Segmented neutrophils, %</td>
<td>63.0 ± 15.5</td>
<td>59.0 ± 15.5</td>
<td>.087</td>
</tr>
<tr>
<td>Band neutrophils, ( \times 10^9, \text{cells/L} )</td>
<td>0.42 ± 0.44</td>
<td>0.83 ± 0.65</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Band neutrophils, %</td>
<td>6.0 ± 6.7</td>
<td>18.3 ± 12.8</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Lymphocytes, ( \times 10^9, \text{cells/L} )</td>
<td>1.4 ± 1.1</td>
<td>0.95 ± 0.92</td>
<td>.001</td>
</tr>
<tr>
<td>Lymphocytes, %</td>
<td>22.4 ± 14.6</td>
<td>19.5 ± 14.5</td>
<td>.14</td>
</tr>
<tr>
<td>Platelets, ( \times 10^9, \text{cells/L} )</td>
<td>213 ± 102</td>
<td>124 ± 69</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>

**NOTE.** Data are ± SD, unless otherwise indicated.
concentrations and the percentage counts were significantly higher in the group of patients with HGE (table 2). The absolute lymphocyte concentration was lower in the patients with HGE than it was in the control patients \(0.95 \times 10^5\) cells/L vs. \(1.40 \times 10^5\) cells/L; \(P < .001\), and although the mean percentage count was lower for the patients with HGE, the difference did not reach statistical significance (table 2).

Further analysis showed an inverse relationship between the mean WBC and Plt concentrations and the risk of having HGE as the cause of the acute nonspecific febrile illness (table 3). Patients with normal WBC or Plt counts were 3–5 times less likely to have HGE than they were to have another febrile illness. In contrast, patients who had a WBC count of \(<2.5 \times 10^9\) cells/L were 5 times more likely to have HGE than were those patients who had a normal WBC count (RR, 5.2; \(P = .047\)). Similarly, Plt counts of \(<100 \times 10^9\) cells/L increased the risk of having HGE more than 10-fold, as was determined by a comparison with patients who had normal Plt counts (RR, 10.2; \(P < .00001\)). Six patients with HGE and 23 control patients had total WBC counts above normal. Four of the control patients had Plt counts above normal, whereas none of the patients with HGE manifested thrombocytosis. The estimated negative predictive values of finding leukocytosis and thrombocytosis in patients who presented with nonspecific fever were 0.793 and 1.000, respectively.

### DISCUSSION

More than 600 cases of HGE have been reported to the Centers for Disease Control and Prevention (Atlanta) since the first case was diagnosed in 1990 (J. Childs, personal communication). The incidence rate for HGE has been estimated to range from <1 to 64 cases per 100,000 population on the basis of retrospective analysis of reported cases from the upper Midwest [8]. Bakken et al. [16] recently reported an HGE seroprevalence rate of 14.9% among permanent residents in northwestern Wisconsin. The limited number of cases of HGE reported to state health agencies to date might suggest that the illness is either underreported or underrecognized.

It has been difficult to predict the severity of illness in individual patients during the early phase of infection. However, the severity of HGE appears to be directly associated with delayed diagnosis and onset of therapy [17–19]. Even though HGE probably is a mild or even subclinical illness in many, if not most, patients [16], at least 7 deaths have been directly or indirectly associated with acute infection [8, 19–21] (Bakken, unpublished data). Therefore, improved understanding of laboratory abnormalities that enable clinicians to identify patients with presumed or probable HGE in the acute care setting is needed, because confirmatory diagnostic laboratory test results may either be delayed or not readily obtainable.

The retrospective study design imposes some limitations on the conclusions that can be drawn from the present investigation. Several characteristic findings were nevertheless noted in the serial mean blood counts. Specific patterns of change in the CBC may help physicians identify patients who should be treated empirically and evaluated further for presumed HGE. This investigation and other published reports demonstrate that patients who have HGE frequently have reduced concentrations of total WBC, Plt, or both [1, 7–11, 14, 19]. However, cytopenias are observed with many nonspecific febrile illnesses, and the mere presence of leukopenia or thrombocytopenia does not permit the clinician to draw specific diagnostic conclusions. Careful examination of a Wright-stained peripheral blood

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**Table 3. Risk of human granulocytic ehrlichiosis (HGE), as determined by the total WBC and platelet counts for patients who present with a nonspecific febrile illness and a history of recent tick exposure or tick bite.**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>No. of control patients ((n = 111))</th>
<th>No. of patients with HGE ((n = 111))</th>
<th>RR</th>
<th>95% CI</th>
<th>(P)</th>
</tr>
</thead>
<tbody>
<tr>
<td>WBC count</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&gt;10.2 (\times 10^9) cells/L</td>
<td>23</td>
<td>6</td>
<td>0.27</td>
<td>0.10–0.71</td>
<td>.010</td>
</tr>
<tr>
<td>4.01–10.2 (\times 10^9) cells/L</td>
<td>68</td>
<td>65</td>
<td>1.00</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>2.5–4.0 (\times 10^9) cells/L</td>
<td>18</td>
<td>30</td>
<td>1.74</td>
<td>0.88–3.42</td>
<td>.156</td>
</tr>
<tr>
<td>&lt;2.5 (\times 10^9) cells/L</td>
<td>2</td>
<td>10</td>
<td>5.23</td>
<td>1.10–24.8</td>
<td>.047</td>
</tr>
<tr>
<td>Platelet count</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&gt;400 (\times 10^9) cells/L</td>
<td>4</td>
<td>0</td>
<td>0.00</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>151–400 (\times 10^9) cells/L</td>
<td>73</td>
<td>36</td>
<td>1.00</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>100–150 (\times 10^9) cells/L</td>
<td>24</td>
<td>24</td>
<td>2.03</td>
<td>1.01–9.05</td>
<td>.067</td>
</tr>
<tr>
<td>&lt;100 (\times 10^9) cells/L</td>
<td>10</td>
<td>51</td>
<td>10.3</td>
<td>4.70–22.7</td>
<td>&lt;.00001</td>
</tr>
</tbody>
</table>

* Not statistically significant at \(P < .05\).
Hematologic Counts and Ehrlichiosis

smear or buffy coat smear done during the acute phase of illness may reveal morulae in granulocytes, a finding diagnostic of active HGE. However, only 25%–70% of reported patients had detectable morulae in the peripheral blood smear [8, 9], and evaluation of the blood smear for morulae remains a test with high specificity but low sensitivity [8].

Although the total WBC counts fell either toward the lower end of or below the reference range during the first week (figure 1), only 76 patients (52.8%) had leukopenia on any given day during the first 2 weeks of illness (figure 2). Consequently, physicians should not exclude the diagnosis of HGE on the basis of a normal total WBC concentration. Thrombocytopenia was noted frequently during the first 8 days of illness (figure 1), and many patients manifested marked thrombocytopenia with individual Plt counts that were as low as cells/915 L. A total of 112 patients (77.8%) displayed thrombocytopenia on 1 or more days, and thrombocytopenia occurred more frequently than did leukopenia during the first 2 weeks of active HGE (P < .0001). Therefore, patients who present with a nonspecific febrile illness of <2 weeks’ duration, a recent history of tick exposure or a tick bite, and a Plt count below the normal range should be considered for empiric antibiotic treatment for HGE and should undergo confirmatory laboratory blood testing. In contrast, patients who present with nonspecific fever of ≥7 days’ duration and either leukocytosis or thrombocytosis have a low probability of having HGE as the cause of their fever (table 3).

Hemoglobin concentrations decreased progressively during the first 2 weeks of illness (figure 1). Not unexpectedly, the mean hemoglobin values were lower among women than they were among men for each day of observation, and the rates of reduction in hemoglobin concentrations versus time were significant and almost parallel for both sexes. The changes in mean hemoglobin concentrations occurred slowly, and no useful patterns that could raise suspicion toward HGE were detected.

The mean values for the relative percentage distribution and absolute concentrations of individual leukocyte cell lines demonstrated several dynamic patterns of change during the observation period. During the first week of illness, the absolute concentrations of band neutrophil leukocytes were significantly elevated (figure 3), and there was a corresponding increase in the mean relative proportions of band neutrophils (figure 4). Individual relative band neutrophil counts that were as high as 56% of total leukocytes were detected during the first week.

Even though the elevated absolute concentrations reverted toward the normal range during the second week of illness (P = .010), there was a sustained release of band neutrophils during the entire observation period (figures 3 and 4). HGE may sometimes be misdiagnosed as an acute viral syndrome because signs and symptoms associated with both illnesses are nonspecific. The finding of increased band neutrophil concentrations should lead to suspicion of HGE or another serious bacterial infection rather than of a viral syndrome, because significantly increased band neutrophil counts are usually not associated with viral illnesses.

Monocytes, eosinophils, and basophils were rarely detected during the acute phase of illness, and low concentrations or the absence of these cell types in peripheral blood samples obtained from a patient who presents with an acute febrile illness should raise the suspicion of HGE. The absolute concentrations of lymphocytes were also markedly reduced during the first few days of observation, even though the lymphocyte percentage count never fell below the reference range (figures 3 and 4). Toward the end of the first week, there was a significant and rapid increase in the lymphocyte concentrations (P < .0001), which continued into the second week. Some patients manifested relative lymphocyte percentage counts as high as 60% during the observation period. Previous investigations have described increased concentrations of γ/δ T lymphocyte subpopulations during the recovery phase of patients who have been treated with doxycycline for human monocytic ehrlichiosis, an infectious disease that is clinically indistinguishable from HGE [22]. These cells appeared as large reactive lymphocytes in the peripheral blood smear. No formal immunophenotypic testing for lymphocyte subtypes was made for the patients that we studied. However, 67% of the patients from New York displayed reactive atypical lymphocytes in peripheral blood smears during the second week of illness, as described in a previous report [9].

In conclusion, the CBC can provide valuable information that may aid physicians in the identification of patients who may be acutely infected with HGE. It is important to understand the dynamic variations and relative changes that occur among the cellular components of the CBC during the period of clinical illness. Normal total concentrations of WBC, Plt, or both, or the failure to recognize morulae in neutrophils by means of microscopic examination of Wright-stained blood smears or buffy coat smears, should not dissuade the clinician from considering HGE as a possible diagnosis in patients who present with a recent history of exposure to ticks and a nonspecific febrile illness. However, significantly reduced WBC or Plt counts increase the likelihood that the patient has acute HGE.

It is important to emphasize that automated differential counting of leukocytes will not be able to differentiate segmented and band neutrophils, and morulae will be missed entirely. Therefore, blood samples from patients who are suspected to have HGE must be evaluated with a manual differential leukocyte count, which also will permit detection of morulae. The diagnostic suspicion of HGE should be heightened by specific patterns that may be displayed by the relative distribution and absolute concentrations of leukocytes. Such changes include increases in the absolute concentration of band
neutrophils combined with marked reductions in the absolute concentrations of lymphocytes and monocytes, and paucity in eosinophils and basophils. Important changes in the differential count include a marked increase in the proportion of band neutrophils, relative lymphopenia, and reduced proportions or absence of monocytes, eosinophils, or basophils. It is important to identify presumptive cases of HGE in the early phase of illness so that empiric treatment with doxycycline or another suitable antibiotic can be initiated in a timely manner to ensure an uncomplicated recovery.

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References