

# Toward the Early Diagnosis of Neonatal Sepsis and Sepsis-Like Illness Using Novel Heart Rate Analysis

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**ABSTRACT.** *Background and Objective.* Abrupt clinical deterioration because of sepsis is a major cause of morbidity and mortality in neonates, and earlier diagnosis should improve therapy of this potentially catastrophic illness. In practice, clinical signs and laboratory data have not been perceived as sensitive or specific for early stages of sepsis. Because heart rate characteristics (HRC) are abnormal during fetal distress and neonatal illness, we hypothesized that abnormal HRC might precede the clinical diagnosis of neonatal sepsis, adding independent information to standard clinical parameters.

*Methods.* In the neonatal intensive care unit at the University of Virginia, we prospectively studied infants admitted from August 1995 to April 1999 who were at risk for developing sepsis. Infants in the sepsis (culture-positive) and sepsis-like illness (culture-negative) groups had an abrupt clinical deterioration that raised clinical suspicion of infection and prompted physicians to obtain blood cultures and start antibiotic therapy. Infants without sepsis raised no clinical suspicion of illness and had no cultures obtained. We measured novel characteristics—moments and percentiles—of normalized heart rate (HR) time series for 5 days before and 3 days after sepsis, sepsis-like illness, or a random time in controls. We also calculated the Score for Neonatal Acute Physiology (SNAP) and the Neonatal Therapeutic Intervention Scoring System (NTISS) as clinical scores of the severity of illness.

*Results.* There were 46 episodes of culture-positive sepsis in 40 patients and 27 episodes of culture-negative sepsis-like illness in 23 patients. We analyzed 29 control periods in 26 patients. Infants with sepsis and sepsis-like illness had lower birth weights and gestational ages and higher SNAP and NTISS scores than did infants without sepsis. The most important new finding was that the infants in the sepsis and sepsis-like illness groups had increasingly abnormal HRC for up to 24 hours preceding their abrupt clinical deterioration. The abnormal HRC were reduced baseline variability and short-lived decelerations in HR. These abnormalities led to significant changes in HRC measures, for example, the third moment (skewness:  $.59 \pm .10$  for sepsis and  $.51 \pm .12$  for sepsis-like illness, compared with  $-.10 \pm .13$  for control over the 6 hours before abrupt deterioration). Culture-positive and culture-negative patients had similar HRC

and clinical scores, including a significant rise in SNAP in the 24 hours before the event. Multivariable logistic regression analysis showed that HRC and clinical scores independently added information in distinguishing infants with sepsis and sepsis-like illness from control patients in the 24 hours before abrupt deterioration.

*Conclusions.* Newborn infants who had abrupt clinical deterioration as a result of sepsis and sepsis-like illness had abnormal HRC and SNAP that worsened over 24 hours before the clinical suspicion of sepsis. A strategy for monitoring these parameters in infants at risk for sepsis and sepsis-like illness might lead to earlier diagnosis and more effective therapy. *Pediatrics* 2001;107:97–104; *heart rate variability, neonatal sepsis, Score for Neonatal Acute Physiology, Neonatal Therapeutic Intervention Scoring System, newborn.*

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ABBREVIATIONS. NICU, neonatal intensive care unit; HRC, heart rate characteristics; HR, heart rate; RR interval, interval between heartbeats; SNAP, Score for Neonatal Acute Physiology; NTISS, Neonatal Therapeutic Intervention Scoring System; SD, standard deviation; p50, 50th percentile data point; ANOVA, analysis of variance; ROC, receiver operating characteristic; SEM, standard error of the mean; CRASH, Cultures, Resuscitation, and Antibiotics Started Here; IL, interleukin.

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**A**brupt clinical deterioration of premature low birth weight infants in the neonatal intensive care unit (NICU) is a common and potentially catastrophic event. Because sepsis is high on the list of possible causes, these episodes prompt physicians to obtain a blood culture and to start antibiotic therapy. Some episodes are the result of culture-proven sepsis, while other episodes that are equally suspicious for sepsis do not have an associated positive blood culture. Some of the sepsis-like episodes with negative blood cultures may be caused by true sepsis with a false-negative blood culture, while others may be initiated by factors such as acute pulmonary or gastrointestinal diseases.

Sepsis and sepsis-like illnesses may be initiated by cytokines and may have a subclinical phase where early detection could be beneficial. For example, Kuster and colleagues<sup>1</sup> have recently found elevated levels of circulating cytokines for up to 2 days before the clinical diagnosis of neonatal sepsis. Early detection of sepsis and sepsis-like illnesses in newborn infants, however, remains difficult using clinical signs and currently available laboratory tests. Clearly, improved early diagnosis of neonatal sepsis and sepsis-like illness is an important goal, and new approaches are required.

We devised a system to examine heart rate char-

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This work was presented in part at the meeting of the Pediatric Academic Societies; May 4, 1999; San Francisco, CA.

Received for publication Dec 15, 1999; accepted Mar 31, 2000.

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acteristics (HRC) for infants in the NICU and used this to test the hypothesis that HRC are altered early in the course of sepsis and sepsis-like illnesses. Because heart rate (HR) patterns are altered during fetal distress and severe neonatal illness,<sup>2-5</sup> we hypothesized that changes in HRC might occur during the early, subclinical stage of illness. In particular, we hypothesized that early stages of sepsis might lead to reduced HR variability and transient HR decelerations, and we developed statistical measures to quantify these changes. Our analytical approach was based on the observation that these abnormal HR patterns lead to asymmetry of frequency histograms of RR intervals, and we used the moments and percentiles of RR distributions to report on this asymmetry.

Thus, this study was undertaken to evaluate HRC in infants at risk for sepsis and sepsis-like illness. To relate these HRC abnormalities to the level of clinical illness, we measured the Score for Neonatal Acute Physiology (SNAP) and the Neonatal Therapeutic Intervention Scoring System (NTISS). The purpose was to determine whether HRC abnormalities preceded an abrupt clinical deterioration that prompted obtaining a blood culture and initiation of antibiotics in infants with sepsis or a sepsis-like illness.

## METHODS

### Study Population

We monitored infants who had risk factors for acquiring late-onset sepsis in the NICU at the University of Virginia from August 1995 to April 1999. These risk factors included low birth weight, prematurity, need for central venous access, and NICU stay >2 weeks. We studied 3 groups of infants defined by the actions of the physicians and the results of blood cultures. Infants who had an abrupt clinical deterioration after 3 days of age that prompted physicians to obtain blood cultures and to give antibiotics were in the sepsis (positive blood culture) or sepsis-like illness (negative blood culture) group. The infants without sepsis group raised no clinical suspicion of sepsis and had no blood cultures obtained over a 10-day period. HRC monitoring results were not visible to the treating physicians and did not influence medical management.

### HR Data Acquisition and Analysis

An analog electrocardiogram voltage signal from the bedside monitor (Marquette, Milwaukee, WI) was digitized and filtered and then evaluated for QRS complexes using specially-equipped personal computers (National Instruments AT-DSP2200, Austin, TX). RR intervals were compared with the previous 100 intervals and excluded if they differed by >5 standard deviations (SDs). All datasets were visually inspected, and records with obviously artifactual data were excluded. This resulted in removal of 1% of all the datasets. HRC measures were calculated from 4096-beat epochs of RR intervals.

### HRC Analysis

We selected HRC measures that give a description of the symmetry of the histogram of RR intervals. Moments are descriptive statistics calculated from the individual differences of data points from the mean. The first moment is itself the mean and the second moment is the SD, the square root of the average of the squared individual differences. The third moment or skewness reports on the symmetry of the histogram. A symmetrical histogram has a skewness of 0, and a histogram with a tail of values that are larger than the median has positive skewness.<sup>6</sup> We also calculated percentiles of the data. The median is the 50th percentile data point (p50), meaning that it resides at the midpoint of the data after sorting from smallest to largest. In addition to the median, which we called the p50, we also calculated the p10, p25, p75, and p90.

Before these calculations, the mean and SD were used to normalize the data, so that the mean and SD of each 4096-beat record were 0 and 1, respectively. This normalization allowed direct comparison of HRC among all records. It is important to note that these measures are based only on the distribution of the data points and not the sequence in which they occur. These measures are not changed by missing points, unlike frequency domain measures.<sup>7</sup>

### Clinical Scores

The SNAP<sup>8</sup> and NTISS<sup>9</sup> scores were obtained either prospectively or from chart review by trained research assistants who were closely supervised (M.P.G.). Scores were calculated for 24-hour epochs relative to the time that the suspicion of sepsis was raised. The investigators were blinded to the results of the HRC analysis at the time of scoring.

### Strategy

To examine the time course of HRC early in sepsis, we analyzed the 5 days before and 3 days after a reference time point. For this, we used the time that the blood culture was obtained (sepsis and sepsis-like illness groups) or a random time (infants without sepsis). The event time for infants without sepsis was assigned randomly during the sixth or seventh day of their 10-day course. We analyzed the data in 6-hour epochs based on this reference point. We calculated HRC for all the 4096-beat datasets and summarized each 6-hour epoch as the median value of each measure for each patient. We excluded epochs with <50% of the expected number of heartbeats. For comparison with clinical illness severity scores in the regression analysis, we analyzed HRC in 24-hour epochs and compared the results with clinical scores obtained over the same period.

### Statistical Analysis

The significance of differences in demographic characteristics and HRC for isolated time points was examined using the Mann-Whitney *U* rank-sum test or analysis of variance (ANOVA; SigmaStat, Jandel, San Rafael, CA). The significance of differences between groups for the 24 hours before and after the event were analyzed using ANOVA with a Tukey test for multiple comparisons (SigmaStat, Jandel). Receiver operating characteristic (ROC) analysis was performed using Microsoft Excel (Microsoft Corporation, Redmond, WA) and the Analyze-it plug-in (Analyze-it Software, Leeds, United Kingdom). We used multivariable logistic regression analysis to examine the ability of HRC and clinical scores to distinguish septic infants from infants without sepsis (S-Plus, MathSoft, Seattle, WA).

## RESULTS

### Study Population

Table 1 shows the demographic characteristics of the infants studied. There were 46 culture-proven episodes of sepsis in 40 patients. There were 2 deaths associated with *Staphylococcus aureus* and *Enterococcus* infection. The most common organisms isolated were coagulase-negative *Staphylococcus* ( $n = 20$ ) and *S aureus* ( $n = 15$ ). There were 27 episodes of culture-negative sepsis in 23 infants. In the control group, there were 29 control periods in 26 patients. In the culture-positive sepsis group, the mean (SD) birth weight, gestational age, and postconceptional age at event were 784 g (409), 26 weeks (2), and 31 weeks (4). In the culture-negative sepsis-like illness group, the values were 756 g (258), 26 weeks (2), and 30 weeks (3). In the infants without sepsis, the values were 976 g (265), 28 weeks (3), and 33 weeks (3). For each parameter, the values for the control group were significantly higher than for the sepsis and sepsis-like illness groups ( $P < .001$ ), but the sepsis group was not significantly different from the sepsis-like illness group.

**TABLE 1.** Population Characteristics

	No Sepsis ( <i>n</i> = 26/29)	Sepsis ( <i>n</i> = 40/46)	Sepsis-Like Illness ( <i>n</i> = 23/27)
Birth weight, g			
<750	3	20	12
750–999	12	14	8
1000–1499	10	5	2
≥1500	1	1	1
Gestational age, wk			
<26	6	19	9
26–28	10	16	9
29–32	9	4	5
>32	1	1	0
Postconceptional age at event, wk			
<26	0	1	4
26–28	0	13	7
29–32	13	20	8
>32	16	12	8
Male sex	15	21	13
White	21	30	21

Data are the number of patients for birth weight and gestational age, and numbers of episodes for postconceptional age at event. The totals are given as (*n* = number of patients/number of episodes) in the column headings.

### An Example of the HR Analysis

Figure 1A shows a time series of 4096 RR intervals from an infant 6 days before an episode of culture-positive sepsis and represents a normal HR pattern. Figure 1, B and C show abnormal RR interval time series from the same infant that were each obtained within 3 to 6 hours before sepsis was suspected and blood cultures were obtained. Both have a baseline of reduced variability and are punctuated by sharp upward deflections that represent short-lived episodes of HR decelerations. Although episodes of bradycardia in NICU patients are common and not necessarily significant,<sup>10</sup> frequent episodes of apnea, which are often associated with HR decelerations, are often interpreted as reflecting early stages of sepsis.<sup>11</sup> The HR in these records always exceeded 120 beats per minute, and these episodes would have failed to trigger HR alarms set at usual thresholds of 100 beats

per minute. In fact, the mean RR intervals of these records are not very different at 323 (1A), 308 (1B), and 302 msec (1C). Distinguishing the data in Fig 1B from normal is nonetheless straightforward by calculating the SD, here 11 msec, compared with 32 msec for the normal. This measure, however, would fail to diagnose the abnormal time series in Fig 1C, where the episodes of subclinical HR decelerations are sufficient to elevate the SD to an apparently normal value of 26 msec.

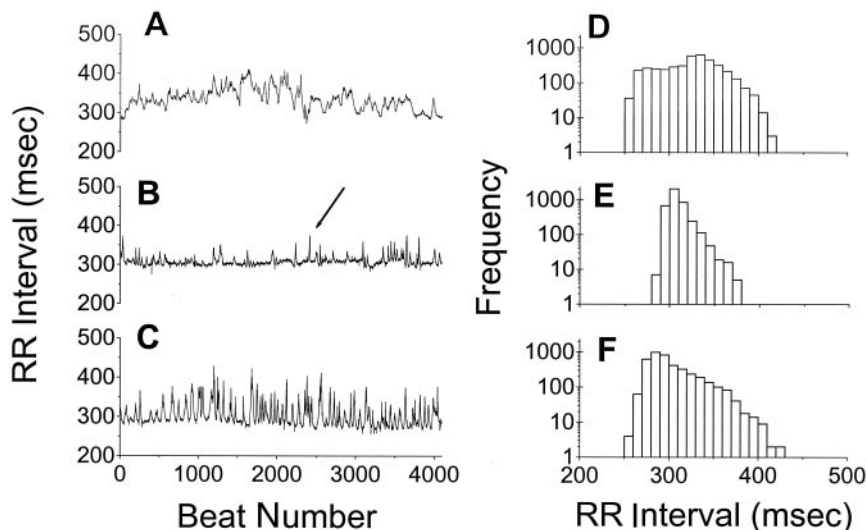
To diagnose these abnormalities, we developed an approach based on the frequency histograms of the RR intervals shown in the right-hand column in Fig 1. The long RR intervals during the decelerations generated asymmetry of the histogram (Fig 1, E and F). We quantified the symmetry of histograms using the third moment or skewness, a descriptive statistic that, like the SD, is based on the differences between individual data points and the mean. The skewness is positive when there is a longer tail of values extending toward longer RR intervals.<sup>6</sup> The skewness values of the 3 time series are different:  $-0.12$  for the data in 1A, indicating a near-symmetric distribution, but 1.99 and 1.33 for the data in Fig 1, B and C, indicating a large degree of asymmetry.

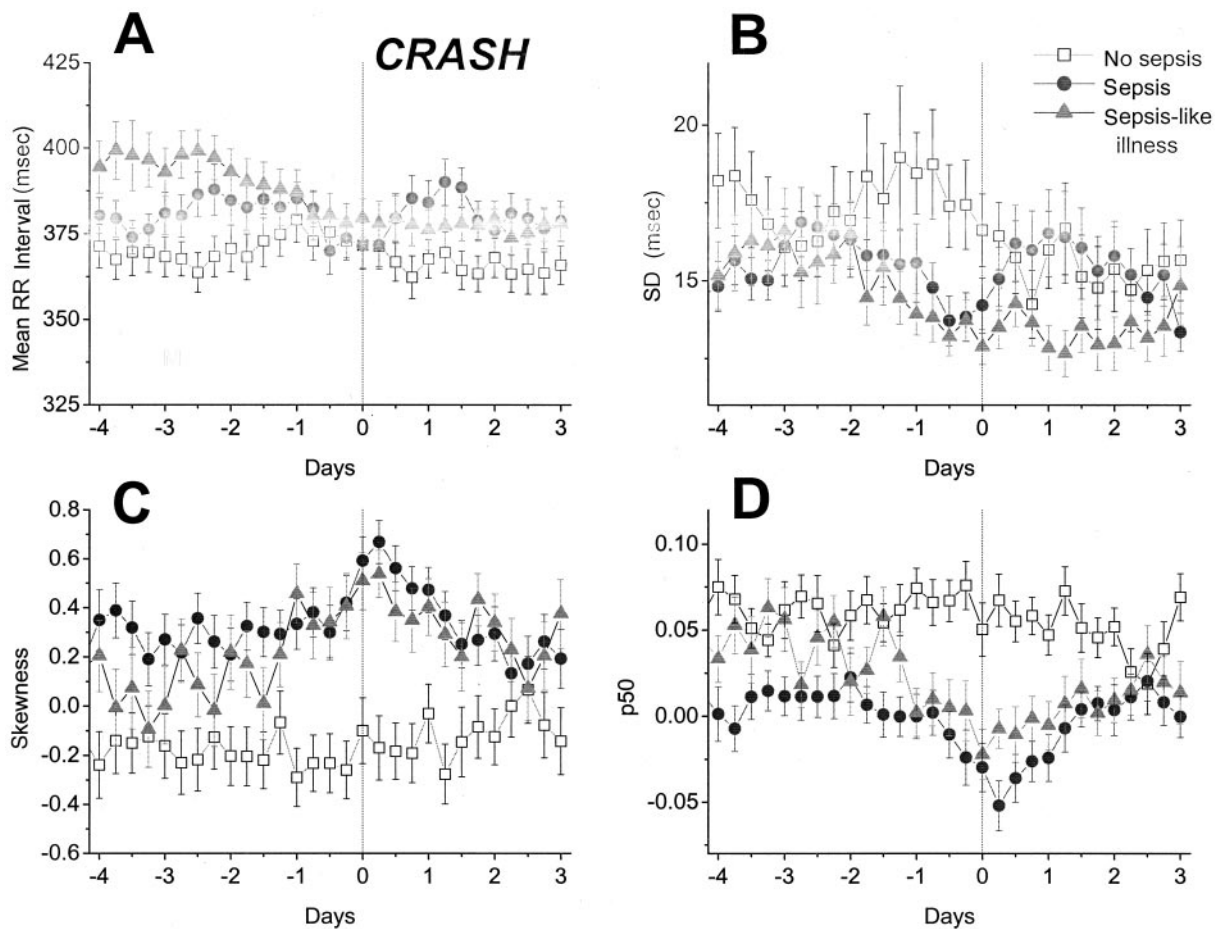
The abnormalities of the histograms can also be quantified by considering the relationship of values in the distribution to the mean. Accordingly, we determined the values of 5 percentile values of the normalized data—the 10th, 25th, 50th (median), 75th, and 90th. These parameters differed among the 3 datasets shown. For example, the p50 was more negative in the abnormal datasets. The values were  $.13$  in the normal record in Fig 1A but  $-.21$  and  $-.33$  in the abnormal records shown in Fig 1, B and C. This change results from the preponderance of values to the left of the center of the distribution, a consequence of the asymmetry of the histogram.

### Summary HRC Data

Figure 2 shows the time course of HRC measures for the 3 patient groups. Each data point is the mean

**Fig 1.** An example of the HRC analysis. A–C, Three 4096-beat RR interval time series and, to the right, their frequency histograms (D–F; note the logarithmic ordinate) are shown. All were recorded from the same infant who had an abrupt clinical deterioration because of coagulase-negative staphylococcal septicemia and an enterococcal urinary tract infection. A shows a normal HR time series recorded 6 days before the event. B and C show abnormal HR time series recorded within 3 to 6 hours before the clinical suspicion of sepsis. The abnormalities are reduced baseline variability and short-lived decelerations of HR. A representative deceleration is marked by an arrow in B. The *y*-axis is RR interval, and the upward spike represents longer RR intervals and thus a slower rate. The data in C show many such decelerations. These changes lead to asymmetry of the frequency histograms with positive skewness; that is, there is a longer tail extending toward higher values of RR intervals.





**Fig 2.** Time course of HRC measures. Data points summarize a 6-hour epoch ending at the time value on the abscissa. Day 0, which ends at the time of the abrupt clinical deterioration, is marked with a vertical line and the word CRASH. Bars are SEM. There are no significant differences in the mean RR interval and SD (A and B). The novel HRC measures skewness and p50, change over the 24 hours before the event in the sepsis and sepsis-like illness groups (C and D).

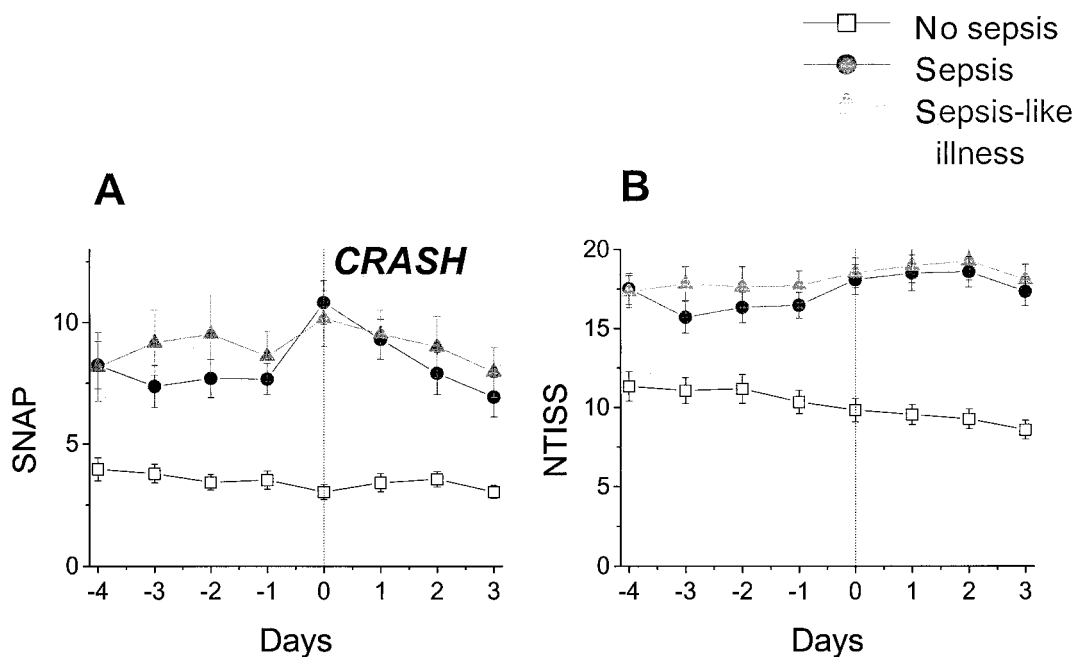
of the median values for 6 hours, and bars are standard error of the mean (SEM). The vertical line marks the time of the abrupt clinical deterioration for which blood cultures were obtained and antibiotics were started. The label CRASH stands for Cultures, Resuscitation, and Antibiotics Started Here. The data points at time 0 represent the 6-hour epoch before, but not including, the time of the deterioration. Values of mean RR interval (Fig 2A) and SD (Fig 2B) did not discriminate among the groups. The skewness (Fig 2C) and p50 values (Fig 2D), in contrast, changed markedly in the epochs from 24 hours before to 24 hours after diagnosis. The skewness was  $.59 \pm .10$  for the sepsis group and  $.51 \pm .12$  for the sepsis-like illness group, compared with  $-.10 \pm .13$  for control over the 6 hours before clinical suspicion. The values for sepsis (culture-positive) and sepsis-like illness (culture-negative) infants were not significantly different, but both were different from the control values ( $P < .001$ ). The p50 was  $-.0298 \pm .014$  for sepsis and  $-.0223 \pm .015$  for sepsis-like illness, compared with  $+.0503 \pm .016$  for control over the 6 hours before clinical suspicion. Again, the values for sepsis and sepsis-like illness groups were not significantly different, but both were different from the control values ( $P < .001$ ).

### Clinical Scores

Figure 3 shows the SNAP and NTISS scores. The sepsis and sepsis-like illness groups had higher levels of both scores throughout, suggesting a greater degree of illness and intensity of interventions. The NTISS level declined over the 10-day period in the control group, consistent with declining requirement for therapies and gradual weaning from support systems. On day 0, the 24 hours before (but not including) the clinical suspicion of sepsis, SNAP rose further in the sepsis and sepsis-like illness groups ( $P = .01$ , ANOVA) and there was no decline in NTISS. Over the 24 hours before and after the CRASH, the values of SNAP and NTISS for sepsis (culture-positive) and sepsis-like illness (culture-negative) infants were not significantly different, but both were different from the control values ( $P < .001$ ).

### Multivariable Logistic Regression Analysis

We performed multivariable logistic regression analysis on the data from the 24 hours before the clinical suspicion of sepsis. HRC were represented by the median values of each of the 5 percentiles, and clinical data were represented by both the SNAP and NTISS scores for this period. We pooled the data from the sepsis and sepsis-like illness groups and



**Fig 3.** Time course of clinical scores. Data points are the mean score over 24 hours; bars are SEM. Both SNAP and NTISS are higher in the sepsis and sepsis-like illness groups. In the 24 hours before diagnosis, SNAP rises further, while NTISS fails to fall.

tested whether a regression model could distinguish the pooled data from the control group. The finding was that the groups were highly significantly different ( $P < .0001$ ; ROC area: .9). Although no single HR percentile measure made a significantly greater contribution than did the others to the discriminatory ability of the model, SNAP contributed significantly more than NTISS ( $P < .003$ , Wald  $z$  test). Both HRC and clinical scores contributed independently to the final model ( $P = .02$ ). Results were similar when only sepsis or sepsis-like illness infants were analyzed, and for regression models using HR moments rather than percentiles.

#### Dynamic Changes in HRC and Illness Severity Scores Early in the Course of Sepsis

We used ROC analysis to quantify differences among the groups and to examine the time course of the differences. The area under the ROC plot is .5 when the groups are not different, and it is 1.0 when the groups are entirely distinct. Panels A and B of Fig 4 show the results for HR moments and percentiles. For this analysis, we show the results after pooling data from the sepsis and sepsis-like groups, but the results for either group individually were very similar. Although mean and SD did not discriminate controls from sepsis and sepsis-like illness infants, skewness showed dynamic differences beginning 12 to 24 hours before the diagnosis. For example, there was a significant difference between the values of skewness measured in the sepsis groups 3 days before sepsis, compared with values 6 to 12 hours before ( $P < .05$ ). The difference then resolved over the 2 days after diagnosis and antibiotic therapy. Two of the HR percentiles, p10 and p50, showed a similar course. SNAP and NTISS also discriminated between the control group and the sepsis and sepsis-like illness groups, and SNAP showed an increasing

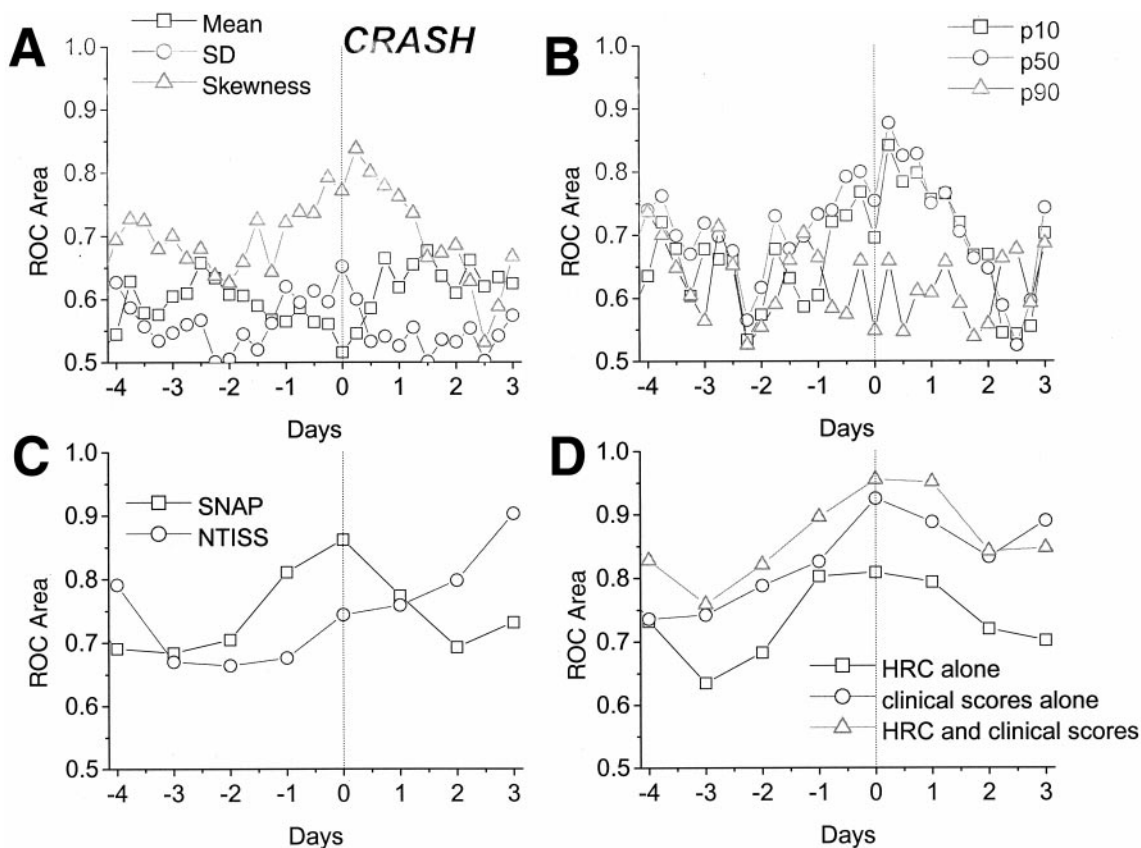
difference for 1 to 2 days before the diagnosis. The increasing ROC area for NTISS is caused by the failure of NTISS to decline in the sepsis and sepsis-like illness infants shown in Fig 3B.

We also used multivariable logistic regression models to distinguish the groups. As shown in Fig 4D, models that used HRC alone and clinical scores (SNAP and NTISS together) alone effectively discriminated the groups, with larger changes at least 1 day before the diagnosis. A model using both HRC and clinical scores was superior, in keeping with the finding above that both HRC and clinical data contributed independently to the model. There were dynamic changes in the regression model values. A comparison of regression model values for day -3 with subsequent days showed that the increases on day 0 were statistically significant for all models ( $P < .05$ ).

#### DISCUSSION

We studied the time course of HRC and clinical illness severity scores in infants at risk for late-onset sepsis in the NICU of a tertiary care hospital. Our most important finding is that abrupt clinical deteriorations that prompted physicians to obtain blood cultures and start antibiotics were preceded for up to 24 hours by increasingly abnormal HRC of reduced baseline variability and subclinical, short-lived decelerations in HR, and by increasingly abnormal SNAP scores.

Abnormalities of HR including reduced beat-to-beat variability and episodes of bradycardia have long been recognized in fetal and neonatal distress.<sup>3-5</sup> These abnormalities should lead to asymmetry of frequency histograms of RR intervals. In this study, we found that the third moment (skewness) and the 10th, 25th, 50th, 75th, and 90th percentile values of normalized RR distributions distinguished



**Fig 4.** Time course of ROC areas (A and B). HRC measures: (C) clinical scores and (D) regression models combining HRC and clinical scores.

records of infants without sepsis from those in early stages of sepsis or sepsis-like illness. Thus, these novel measures serve to quantify well-established markers of early fetal and neonatal distress, and they may add to clinical observations by detecting sub-clinical changes in HRC. In addition, the new measures have the advantage of reliability in datasets with missed beats, unlike conventional frequency domain measures of HR time series.<sup>7,12</sup>

Early diagnosis of neonatal sepsis is difficult,<sup>13</sup> because the clinical signs are neither uniform nor specific. One limitation of our study is that we did not standardize the criteria for obtaining blood cultures and giving antibiotics, but rather observed the practices of the physicians. The threshold for obtaining blood cultures and giving antibiotics differs among physicians, and our study groups may have been heterogeneous for level of clinical illness as a result. The rise in SNAP for the 24 hours before the CRASH, however, confirms that there was worsening of physiologic variables in the sepsis groups, as expected for infants with clinical deterioration.

Not all patients with clinical signs of sepsis have positive blood cultures. The current hypothesis is that the clinical syndrome is brought about by the host response to insults, such as bacterial infection. The major host response is release of cytokines, small circulating peptides that serve as mediators of the inflammatory response. The syndrome common to sepsis and sepsis-like illness has been named the systemic inflammatory response syndrome,<sup>14</sup> and

the pathogenesis suggested to be an imbalance between proinflammatory and antiinflammatory effects of cytokines.<sup>15</sup>

We do not know the mechanism of the HRC changes. In sepsis and sepsis-like illness, circulating cytokines play a major role in initiating and maintaining the inflammatory response, and cytokine levels correlate with the severity of illness.<sup>16–18</sup> Cytokines have widespread effects on signal transduction processes and may interfere with normal events of HR control by the sympathetic and parasympathetic nervous systems. For example, the cytokines tumor necrosis factor- $\alpha$ , interleukin (IL)-1 $\beta$ , and IL-6 increase HR but blunt HR responses to  $\beta$ -adrenergic agonists.<sup>19,20</sup> In addition, sepsis and sepsis-like illness are associated with alterations in  $\beta$ -adrenergic receptor number and distribution<sup>21,22</sup> and with multiple steps of signal transduction via  $\beta$ -adrenergic receptors.<sup>23</sup> The recent demonstration that circulating levels of the cytokines IL-1 receptor antagonist and IL-6 are elevated for 2 days before clinical evidence of neonatal sepsis and sepsis-like illness<sup>1</sup> is consistent with our finding of changes in HRC and clinical scores 12 to 24 hours before the clinical diagnosis. Interestingly, HR variability is reduced by infusion of endotoxin in human volunteers.<sup>24</sup> More precise delineation of the mechanism of the findings awaits a better understanding of neonatal HR control during illness and health.

Although the cause of illness is not known with certainty in the infants with negative blood cultures,

we suspect that infection was the cause in several. Although the blood culture is believed to be the gold standard for establishing the diagnosis of sepsis caused by systemic bacterial infection, there are concerns regarding its reliability,<sup>25</sup> especially if single samples of small volume are submitted<sup>26,27</sup> as is often the practice in critically ill newborn infants. For example, as many as 60% of culture results may be falsely negative if only .5 mL of blood is obtained from infants with low colony count sepsis.<sup>28</sup> In a study of 298 aerobic culture specimens, the mean blood volume submitted was .53 mL and 55% of samples contained <.5 mL.<sup>29</sup> We might then expect 30% to 40% of all infants with sepsis to have negative blood cultures. This is in agreement with 2 studies in which premortem blood cultures failed to identify ~20% of infants with infection proven by postmortem cultures and autopsy.<sup>30,31</sup>

The findings are limited by differences in birth weight, gestational age, and postconceptional age in the infants without sepsis, compared with the other groups. Infants who developed sepsis and sepsis-like illness had lower birth weight (~200 g) and gestational age and postconceptional age (~2 weeks) and would be recognized clinically as having increased risk. They also had more abnormal HRC, SNAP, and NTISS at 4 days before the abrupt clinical deterioration, reflecting this baseline difference of increased risk. Our definition required at least 10 consecutive days during which the infant did not receive antibiotics and did not have symptoms suggestive of sepsis that prompted physicians to obtain a blood culture. This eliminated some of the high-risk very low birth weight and gestational age infants, because their increased probability of sepsis and sepsis-like illness frequently led physicians to obtain blood cultures and give antibiotics. Thus, we were unable to match precisely the characteristics of the groups with sepsis and sepsis-like illness.

Importantly, we found dynamic changes of SNAP and HRC for up to 24 hours before the abrupt clinical deterioration. We found, for example, that the SNAP score rose in the 1-day epoch before the abrupt clinical deterioration, suggesting that there is diagnostic information in conventional vital signs and laboratory tests. Because SNAP is calculated only on a 24-hour basis, however, we do not know whether the physiologic and laboratory abnormalities measured by SNAP appeared just before the clinical suspicion of sepsis and sepsis-like illness or whether they occurred gradually over the 24-hour period. We also found significant dynamic changes in novel HRC, such as skewness and p50, that may add to clinical observation in early detection of sepsis and sepsis-like illness. An advantage of HRC over clinical scores is their noninvasive and continuous nature.

Clinicians have found no clinical signs or laboratory test findings to be reliable for very early diagnosis of neonatal sepsis, and 10 to 20 infants are treated for each infant with a positive blood culture.<sup>32</sup> Successful surveillance strategies leading to an earlier diagnosis of sepsis are urgently needed for very low birth weight infants to decrease mortality and morbidity.<sup>33</sup> Our findings suggest that monitor-

ing of HRC might be developed into a useful adjunct to conventional clinical observation and practice. The analysis presented here based on clinical characteristics and HRC is effective in distinguishing low-risk infants having a stable course from high-risk infants hours before the clinical diagnosis of sepsis. A more useful test would be to detect early, subclinical sepsis in high-risk infants whose baseline HRC are not normal. There was a significant difference between the values of skewness measured in the sepsis groups 3 days before sepsis, compared with values 6 to 12 hours before. This finding affirms the dynamic nature of the HRC changes and motivates further research on clinical implementation of HRC monitoring.

## ACKNOWLEDGMENTS

This work was supported by the American Heart Association, Mid-Atlantic Consortium; Children's Medical Center Research Fund, University of Virginia; Virginia's Center for Innovative Technology; and Medical Automation Systems, Charlottesville, Virginia.

We thank T. Smoot, J. Nelson, K. Monahan, J. Richman, A. Dinn, H. Huq, F. Artrip, R. Vaughn, T. Tamburello, W. King, and S. Booth for integral contributions to the data acquisition and analysis; F. Harrell and E. Bissonette for regression analysis; and G. Beller, J. Kattwinkel, and B. Duling for support.

## REFERENCES

1. Kuster H, Weiss M, Willeitner AE, et al. Interleukin-1 receptor antagonist and interleukin-6 for early diagnosis of neonatal sepsis 2 days before clinical manifestation. *Lancet*. 1998;352:1271-1277. See comment
2. Griffin MP, Scollan DF, Moorman JR. The dynamic range of neonatal heart rate variability. *J. Cardiovasc. Electrophysiol*. 1994;5:112-124
3. Burnard ED. Changes in heart size in the dyspnoeic newborn infant. *Br Med J*. 1959;1:1495-1500
4. Rudolph AJ, Vallbona C, Desmond MM. Cardiodynamic studies in the newborn. III. Heart rate patterns in infants with idiopathic respiratory distress syndrome. *Pediatrics*. 1965;36:551-559
5. Cabal LA, Siassi B, Zanini B, Hodgman JE, Hon EE. Factors affecting heart rate variability in preterm infants. *Pediatrics*. 1980;65:50-56
6. Weisstein EW. *CRC Concise Encyclopedia of Mathematics*. Boca Raton, FL: Chapman and Hall/CRC; 1999
7. Berntson GG, Stowell JR. ECG artifacts and heart period variability: don't miss a beat. *Psychophysiology*. 1998;35:127-132
8. Richardson DK, Gray JE, McCormick MC, Workman K, Goldmann DA. Score for neonatal acute physiology: a physiologic severity index for neonatal intensive care. *Pediatrics*. 1993;91:617-623
9. Gray JE, Richardson DK, McCormick MC, Workman-Daniels K, Goldmann DA. Neonatal therapeutic intervention scoring system: a therapy-based severity of illness index. *Pediatrics*. 1992;90:561-567
10. Hodgman JE, Hoppenbrouwers T, Cabal LA. Episodes of bradycardia during early infancy in the term-born and preterm infant. *Am J Dis Child*. 1993;147:960-964
11. Fanaroff AA, Korones SB, Wright LL, et al. Incidence, presenting features, risk factors and significance of late onset septicemia in very low birth weight infants. The National Institute of Child Health and Human Development Neonatal Research Network. *Pediatr Infect Dis J*. 1998;17:593-598
12. Schechtman VL, Kluge KA, Harper RM. Time domain system for assessing variations in heart rate. *Med Biol Engl Comp*. 1988;26:367-373
13. Escobar GJ. The neonatal "sepsis work-up": personal reflections on the development of an evidence-based approach toward newborn infections in a managed care organization. *Pediatrics*. 1999;103:360-373
14. Members of the ACCP/SCCM Consensus Conference Committee. American College of Chest Physicians/Society of Critical Care Medicine Consensus Conference. Definitions for sepsis and organ failure and guidelines for the use of innovative therapies in sepsis. *Crit Care Med*. 1992;20:864-874
15. Bone RC, Grodzin CJ, Balk RA. Sepsis: a new hypothesis for pathogenesis of the disease process. *Chest*. 1997;112:235-243
16. Anderson MR, Blumer JL. Advances in the therapy for sepsis in children. *Pediatr Clin North Am*. 1997;44:179-205

17. Harris MC, Costarino ATJ, Sullivan JS, et al. Cytokine elevations in critically ill infants with sepsis and necrotizing enterocolitis. *J Pediatr*. 1994;124:105-111
18. Glauser MP, Heumann D, Baumgartner JD, Cohen J. Pathogenesis and potential strategies for prevention and treatment of septic shock: an update. *Clin Infect Dis*. 1994;18:S205-S216
19. Oddis CV, Finkel MS. Cytokines and nitric oxide synthase inhibitor as mediators of adrenergic refractoriness in cardiac myocytes. *Eur J Pharmacol*. 1997;320:167-174
20. Oddis CV, Simmons RL, Hattler BG, Finkel MS. Chronotropic effects of cytokines and the nitric oxide synthase inhibitor, L-NMMA, on cardiac myocytes. *Biochem Biophys Res Commun*. 1994;205:992-997
21. Tang C, Yang J, Liu MS. Progressive internalization of  $\beta$ -adrenoceptors in the rat liver during different phases of sepsis. *Biochim Biophys Acta*. 1998;1407:225-233
22. Hahn PY, Yoo P, Ba ZF, Chaudry IH, Wang P. Upregulation of Kupffer cell  $\beta$ -adrenoceptors and cAMP levels during the late stage of sepsis. *Biochim Biophys Acta*. 1998;1404:377-384
23. Bernardin G, Strosberg AD, Bernard A, Mattei M, Marullo S.  $\beta$ -adrenergic receptor-dependent and -independent stimulation of adenylate cyclase is impaired during severe sepsis in humans. *Int Care Med*. 1998;24:1315-1322
24. Godin PJ, Fleisher LA, Eidsath A, et al. Experimental human endotoxemia increases cardiac regularity: results from a prospective, randomized, crossover trial. *Crit Care Med*. 1996;24:1117-1124
25. Kaftan H, Kinney JS. Early onset neonatal bacterial infections. *Semin Perinatol*. 1998;22:15-24
26. Aronson MD, Bor DH. Blood cultures. *Ann Intern Med*. 1987;106:246-253
27. Kellogg JA, Ferrentino FL, Goodstein MH, Liss J, Shapiro SL, Bankert DA. Frequency of low level bacteremia in infants from birth to two months of age. *Pediatr Infect Dis J*. 1997;16:381-385
28. Schelonka RL, Chai MK, Yoder BA, Hensley D, Brockett RM, Ascher DP. Volume of blood required to detect common neonatal pathogens. *J Pediatr*. 1996;129:275-278
29. Neal PR, Kleiman MB, Reynolds JK, Allen SD, Lemons JA, Yu PL. Volume of blood submitted for culture from neonates. *J Clin Microbiol*. 1986;24:353-356
30. Pierce JR, Merenstein GB, Stocker JD. Immediate postmortem cultures in an intensive care nursery. *Pediatr Infect Dis*. 1984;3:510-513
31. Squire E, Favara B, Todd J. Diagnosis of neonatal bacterial infection: hematologic and pathologic findings in fatal and nonfatal cases. *Pediatrics*. 1979;64:60-64
32. Gerdes JS, Polin RA. Sepsis screen in neonates with evaluation of plasma fibronectin. *Pediatr Infect Dis J*. 1987;6:443-446
33. Stoll BJ, Gordon T, Korones SB, et al. Late-onset sepsis in very low birth weight neonates: a report from the National Institute of Child Health and Human Development Neonatal Research Network. *J Pediatr*. 1996;129:63-71

... Normal healthy development of the nervous centres demands quiet, rest, peaceful surroundings, and freedom from everything which causes excitement or undue stimulation.

The steadily increasing frequency of functional nervous diseases among young children is one of the most powerful arguments for greater attention by physicians to the subject of the hygiene of the nervous system during infancy. Most parents err through ignorance. Playing with young children, stimulating to laughter and exciting them by sights, sounds, or movements until they shriek with apparent delight, may be a source of amusement to fond parents and admiring spectators, but it is almost invariably an injury to the child. This is especially harmful when done in the evening. It is the plain duty of the physician to enlighten parents on this point and insist that the infant shall be kept quiet, and that all such playing and romping as has been referred to shall, during the first year at least, be absolutely prohibited.

Holt LE. *The Diseases of Infancy and Childhood*. New York, NY: D. Appleton and Company; 1899:5

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