Pain Expression in Neonates: Facial Action and Cry

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Pain expression in neonates: facial action and cry

Ruth V.E. Grunau and Kenneth D. Craig

British Columbia Children's Hospital and University of British Columbia,
Vancouver, B.C. V5Z 1L9 (Canada)

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Summary

Pain expression in neonates instigated by heel-lance for blood sampling purposes was systematically described using measures of facial expression and cry and compared across sleep/waking states and sex. From gate-control theory it was hypothesized that pain behavior would vary with the ongoing functional state of the infant, rather than solely reflecting tissue insult. Awake-alert but inactive infants responded with the most facial activity, consistent with current views that infants in this state are most receptive to environmental stimulation. Infants in quiet sleep showed the least facial reaction and the longest latency to cry. Fundamental frequency of cry was not related to sleep/waking state. This suggested that findings from the cry literature on qualities of pain cry as a reflection of nervous system 'stress,' in unwell newborns, do not generalize directly to healthy infants as a function of state. Sex differences were apparent in speed of response, with boys showing shorter time to cry and to display facial action following heel-lance. The findings of facial action variation across sleep/waking state were interpreted as indicating that the biological and behavioral context of pain events affects behavioral expression, even at the earliest time developmentally, before the opportunity for learned response patterns occurs. Issues raised by the study include the importance of using measurement techniques which are independent of preconceived categories of affective response.

Key words: Neonate; Pain expression; Facial action; Cry

Introduction

Neonatal pain behavior and its corresponding biological substrates provide the foundation for subsequent socialization and the development of pain expression. Pain behavior in adults is multifaceted, reflecting not only tissue damage but also learned coping strategies, socialization and cultural variation [30]. Neonatal pain behavior, in contrast, occurs prior to the possibility of learned reaction patterns.

1 This research is based on a doctoral dissertation submitted to the University of British Columbia by the first author, and was supported by research grants to the second author by the Social Sciences and Humanities Research Council and Natural Sciences and Engineering Research Council of Canada.

Correspondence to: Dr. Ruth V.E. Grunau, Department of Psychology, B.C. Children's Hospital, Vancouver, B.C. V6H 3V4, Canada.
Thus it forms the basis for subsequent maturation and the integration of sensory, cognitive, affective, and behavioral events, within specific familial and cultural contexts, which may mediate the experience and behavioral responses to noxious stimuli in children and adults [10]. Pain expression in infancy is only beginning to receive systematic study and has been described as a neglected field despite its importance for management of pediatric pain [5,23,34].

In order to examine how sensory and behavioral aspects of pain may interrelate in neonates, it may be productive to extrapolate from what is known about adults. From clinical and research evidence in support of Gate Control Theory [30], it is clear that pain behavior in adults is not solely a reflection of degree of tissue damage but, rather, pain expression is strongly affected by the contextual events in which pain occurs. Due to the lack of opportunity in newborns for learned associations with pain events, it would be easy to assume that at the earliest time of infancy pain expression is a direct reflection of degree of tissue damage only. If, on the other hand, pain in neonates were found to vary with situational variables, it would suggest that the underlying biological mechanisms whereby context serves as a powerful moderator of pain expression in adults are already functional at the earliest stage of development.

A powerful, physiologically based contextual variable for newborns is their sleep/waking state. State organization is viewed currently as an indication of the infant's neurobehavioral status, reflecting the infant's ongoing internal organization and capacity for integrating environmental input [16,36]. The construct of sleep/waking state has been found to offer an important organizational network for studying perceptual systems in neonates [37,40], however the tactile modality has received little attention and has mainly addressed differential effects during sleep states only [14,38]. Vlach et al. [42] studied state dependency of non-invasive tactile reflexes in newborns across quiet sleep, active sleep and quiet wakefulness, showing state-related variation in responses to gentle stroking of the extremities, but not for limb girdle reflexes. With invasive rather than non-invasive tactile stimulation, changes in sleep patterns have been found following circumcision [1,21]. The present study compared behavioral changes in response to pain as a function of prior sleep and waking states in order to examine the possibility that sleep/waking state may mediate responsivity to pain in neonates.

To determine whether sex-role stereotypes of females as more responsive to pain have a biological basis we compared the impact of noxious stimuli across sexes in early infancy before socialization begins to take place. Earlier studies of neonatal sex differences in pain-related behavior have yielded inconsistent findings. Female neonates were found to show greater reaction, as compared with males, in behavioral response to an air jet to the abdomen [2,3] and to electrotactual stimulation [29]. On the other hand, Gullickson and Crowell [20] found no sex differences to electrotactual stimulation. Recently, Owens and Todt [35] found no sex differences in response to heel-lance for PKU blood collection. Thus, the question remains open.

Acoustic analysis of cry has been proposed as a potentially important measure in infant pain research [27]. Cry features such as fundamental frequency have not been
productive in distinguishing ‘cry types,’ such as ‘pain cry’ from ‘hunger cry,’ as it appears that cry is a graded signal rather than one which reflects qualitative variation in experience [32]. However, within the study of parameters of pain itself acoustic measures may be usefully. In the literature on cry in unwell as compared with healthy neonates, fundamental frequency of the first cry following pain is currently conceptualized as indicative of the central nervous system capacity for response modulation [18,26]. There appear to be no investigations of variation in cry features as a function of pre-stimulus state, or of responses to invasive procedures such as the heel-lance across state.

Fine grained analysis of facial activity [15] has been used in the study of painful expression in adults [12,13] but not children. Children display ample non-verbal distress during pain [11]. A coding system was developed to examine facial activity in neonates instigated by noxious stimulation.

The aim of this study was to examine facial movement and vocalization to discomfort (heel-rub) and pain (heel-lance) as a function of 4 sleep/waking states (quiet/sleep, active/sleep, quiet/awake, active/awake) and sex. It was hypothesized that higher fundamental frequency of cry would be evident in those states which evoked the most facial reactivity. State differences as mediators of infant responsivity to invasive stimulation could operate in a number of different ways. The contrasting hypotheses were as follows:

1. Response patterns induced by noxious events will differ depending on whether the infant is asleep or awake.

2. The rest–activity cycle which occurs within both sleeping and waking states may be a more important key to ongoing changes than whether the infant appears to be asleep or awake [24,39]. Based on this model, responses will be more similar for quiet/sleep and quiet/awake infants than active/sleep and active/awake infants.

3. Quiet wakefulness has been viewed as reflecting optimal orientation to environmental input [7,8]. This implies that responses in the quiet/awake state will differ from the other states.

Methods

Subjects

A continuous sample of 77 boys and 63 girls from the well-baby unit of a major metropolitan maternity hospital participated in the study. Criteria for inclusion were: birthweight above 2500 g; gestation of 38–42 weeks; Apgar at 5 min of 8–10; circumcision had not taken place. The mean age of the infants was 43.05 h (S.D. = 7.06 h) and mean birthweight 3446.4 g, (S.D. = 425.9 g). Mean maternal age was 29.3 years (range 17–42 years); 89 (63%) of the mothers were white, 38 (27%) Oriental, 8 (6%) East Indian and the remaining 4% comprised 4 Filipino and 1 North American Indian. Mode of delivery was 72 (52%) spontaneous vaginal, 34 (24%) vaginal forceps, 21 (15%) planned cesarian section, and 13 (9%) cesarian section following labor.
Apparatus

A Panasonic WV-3900 color camera was used for video recording with 3/4" video tape. In addition to audio recording on the video tape, separate sound recordings were carried out on Ampex precision 1.5 ml polyester magnetic audio tape on a Sony TC377 reel to reel tape recorder using 1/4" track recording. VU levels were set prior to recording with a 1000 Hz tone. No noise suppression was used, and VU levels were not adjusted during recording to account for peaking. An AKG D109 Lavalier microphone was suspended approximately 18 cm from the infant's mouth. To cue the heel-lance event, an inaudible tone of 1000 Hz was triggered using a shure mixer connected straight into the separate audio system, and to the audio portion of the video system.

The original 3/4" videotapes were copied onto 1/2" VHS tapes with an RCA time-date generator used to superimpose a digital time display for coding purposes. An RCA VHS video selectavision tape recorder (model VET650) with remote control and an RCA 48 cm playback color monitor (JD975WV) were used during video coding.

Procedure

Informed consent was obtained from a parent, usually the mother. Testing was conducted in a quiet room near the nursery, between 7 a.m. and 9 a.m. The infant remained in the bassinet.

Using Prechtl's [36] observational rating system, sleep/waking state [(1) eyes closed, no facial movement (quiet/sleep); (2) eyes closed, facial movement (active/sleep); (3) eyes open, no facial movement (quiet/awake); (4) eyes open, facial movement (active/awake)] was recorded by the first author in the nursery, over a 60 sec period. Infants who were already crying at the time sleep/awake state was recorded, as well as 7 infants who did not cry in response to heel lance, were excluded from the study. To establish interjudge reliability of the state rating, babies also were scored in real time by two blind coders using the first 3 sec of videotape.

Video and sound recording were carried out prior to and during the routine heel-lance procedure, which was performed by a hospital laboratory technician for the purpose of PKU screening of blood samples. The camera provided a close-up view of the infant's face at all times. The baby remained partially swaddled. The lab technician's standard protocol involved: checking the infant's identification band on either the wrist or ankle; picking up the foot and rubbing it to disinfect the skin (heel-rub phase). The heel-lance procedure then involved use of a small disposable metal scalpel (4.9 mm long point microlance) for incision (heel-lance phase). The heel then was squeezed and blood samples were collected on 4 circled areas on an absorbent card. There were 9 lab technicians involved on rotating shifts with the number of infants seen by separate technicians ranging between 4 and 42.

Measures

Acoustic analysis. Cry was defined as 'audible vocalization.' Cries were transferred from the Ampex magnetic tape to a PDP 11/34 digital computer, converted from analog to digital signal and sampled at 20,000/sec. The 20 kHz sampling rate
yielded high wave form resolution. The digitized waveform was displayed on a VF11 vector graphics terminal.

Durational measurements of latency from the end of the tone (signaling the heel-lance event) to the start of the first cry expiration (cry latency), and from the start to the end of the first cry expiration (cry duration), were then determined.

Fundamental frequency (perceived as pitch) is the first harmonic in a complex periodic sound wave [28]. Pitch analysis used short-term autocorrelation methods [31]. Pitch plots of the first cry expiration were displayed on a Tektronix videoscope, then printed. Maximum and minimum fundamental frequencies (f₀) of the first cry expiration were recorded. In addition, fundamental frequency was recorded for the first pitch point (f₀¹) then at 100 msec (f₀²) and 200 msec (f₀³) to examine time-related features of changes in the initial portion of the first cry.

The number of cry cycles from each breath inspiration to expiration over 30 sec from heel-lance was counted independently by two coders from the audiotapes.

*Video analysis*

A coding system for facial movement was based on the FACS approach [12,15] to coding action units representing discrete facial movements. The system represented an adaptation of Oster's system [33] for coding babies' faces. Movement represented

<table>
<thead>
<tr>
<th>TABLE I</th>
<th>NEONATAL FACIAL CODING SYSTEM</th>
</tr>
</thead>
<tbody>
<tr>
<td>Action</td>
<td>Description</td>
</tr>
<tr>
<td>Brow bulge</td>
<td>Bulging, creasing and vertical furrows above and between brows occurring as a result of the lowering and drawing together of the eyebrows.</td>
</tr>
<tr>
<td>Eye squeeze</td>
<td>Identified by the squeezing or bulging of the eyelids. Bulging of the fatty pads about the infant's eyes is pronounced.</td>
</tr>
<tr>
<td>Naso-labial furrow</td>
<td>Primarily manifested by the pulling upwards and deepening of the naso-labial furrow (a line or wrinkle which begins adjacent to the nostril wings and runs down and outwards beyond the lip corners).</td>
</tr>
<tr>
<td>Open lips</td>
<td>Any separation of the lips is scored as open lips.</td>
</tr>
<tr>
<td>Stretch mouth (vertical)</td>
<td>Characterized by a tautness at the lip corners coupled with a pronounced downward pull on the jaw. Often stretch mouth is seen when an already wide open mouth is opened a fraction further by an extra pull at the jaw.</td>
</tr>
<tr>
<td>Stretch mouth (horizontal)</td>
<td>This appears as a distinct horizontal pull at the corners of the mouth.</td>
</tr>
<tr>
<td>Lip purse</td>
<td>The lips appear as if an 'oo' sound is being pronounced.</td>
</tr>
<tr>
<td>Taut tongue</td>
<td>Characterized by a raised, cupped tongue with sharp tensed edges. The first occurrence of taut tongue is usually easy to see, often occurring with a wide open mouth. After this first occurrence, the mouth may close slightly. Taut tongue is still scorable on the basis of the still visible tongue edges.</td>
</tr>
<tr>
<td>Chin quiver</td>
<td>An obvious high frequency up-down motion of the lower jaw.</td>
</tr>
</tbody>
</table>
the key to scoring the actions which are presented in Table I. A coder who had been trained in the adult FACS system [15] and was blind to the purpose of the study and all information about the infants did the scoring. Coding was carried out using a slow motion and stop-frame feedback system. Reliability coding of the discrete facial actions was done by the first author. A second blind coder carried out reliability coding on the state and cry cycle data.

Each face variable, namely brow, eye squeeze, naso-labial furrow, open lips, vertical stretch mouth, horizontal stretch mouth, lip purse, taut tongue and chin quiver was scored if the action occurred. This was done for each of the 9 facial actions separately for each of 12 segments of the heel-lance sequence.

Thirty-six continuous seconds of video tape, divided into 3-sec segments, were scored for each subject. Segments 1 and 2 immediately preceded the heel-lance and showed responses to heel-rub; heel-lance took place in segment 3; during segments 4–12, blood was collected, so these reflect not only the responses to heel-lance, but also to the heel squeezing which occurred in the course of blood collection.

Results

Preliminary analyses

Frequency and percent of occurrence of each of the 9 face variables were examined prior to data analysis. Brow bulge, eye squeeze, naso-labial furrow and open lips were observed in 99% of the infants within 6 sec of heel-lance; taut tongue in 70% of infants and vertical stretch mouth in 43%. In contrast no infant showed horizontal stretch mouth or pursed lips within 6 sec of heel-lance. Thus, these two facial actions were not considered any further.

Chin quiver occurred only slightly more often (6%) so it was not analyzed individually, but was included in total amount of face movement when all actions were summed.

Reliability

Facial coding. Interobserver reliability was 0.88, computed for a randomly selected 20% of the subjects, for each face variable in each segment, using the conservative Facial Action Coding System reliability formula [15]:

\[
\text{coefficient of reliability} = \frac{(\text{number of actions on which coder 1 and coder 2 agreed} \times 2)}{(\text{number of actions scored by the 2 coders})}
\]

Number of cry cycles. Rate of agreement was 85% with an error margin of +1 cry cycle.

State. Inter-rater agreement between the two coders was 83%.

Technician effects

There was no evidence infants in any particular state were assigned disproportionately across technicians, \(\chi^2(24) = 22.02, P = 0.58\). Although technician ef-
effects were not important in analysis of the study hypotheses, they were of interest in their own right. Chi-square analyses examined each facial action for each segment across the 9 lab technicians. For the heel-rub analyses, statistically significant technician effects at \( \alpha = 0.01 \) were obtained for naso-labial furrow in segment 1, \( \chi^2(8) = 21.84, P < 0.005 \), and segment 2, \( \chi^2(8) = 23.52, P < 0.003 \), and for eye squeeze in segment 2, \( \chi^2(8) = 20.53, P < 0.008 \). Thus facial expression reflected technician differences during the heel-rub phase.

Following heel-lance, which occurred in segment 3, there were no significant technician effects in segments 3–7, i.e., for 15 sec after heel-lance. Of the remaining segments, which reflected heel squeezing to complete blood collection, the following significant technician effects were found: brow bulge segment 11, \( \chi^2(8) = 28.43, P < 0.0004 \) and segment 12, \( \chi^2(8) = 22.35, P < 0.0004 \); eye squeeze segment 10, \( \chi^2(8) = 19.75, P < 0.01 \), segment 11, \( \chi^2(8) = 31.08, P < 0.0001 \), segment 12, \( \chi^2(8) = 26.91, P < 0.0007 \); naso-labial furrow segment 11, \( \chi^2(8) = 21.90, P < 0.005 \), segment 12, \( \chi^2(8) = 21.51, P < 0.0009 \); lip part segment 8, \( \chi^2(8) = 30.50, P < 0.0002 \), segment 10, \( \chi^2(8) = 19.69, P < 0.01 \), segment 11, \( \chi^2(8) = 26.87, P < 0.0007 \); vertical stretch mouth segment 10, \( \chi^2(8) = 19.58, P < 0.01 \); taut tongue segment 8, \( \chi^2(8) = 19.13, P < 0.01 \). Technicians clearly had a variable impact on facial expression subsequent to the heel-lance, but only in time segments 8–12. These segments reflected impact of heel-squeezing rather than lancing.

The impact of technicians on cry measures called for univariate one-way analysis of variance (ANOVA) across technicians on \( f_1^b, f_2^b, f_3^b \), number of cry cycles, cry duration and latency to cry. There were no statistically significant technician effects on cry measures \((P > 0.05)\).

**Time since feeding.** Since the PKU blood collection procedure was carried out between 7 a.m. and 9 a.m. only, it was not possible to control for time from feeding. This was a potentially important confounding factor as infant behavioral state may vary with hunger. A one-way ANOVA was performed on time since feeding to heel-lance across sleep/wake states. Mean time since feeding for all subjects was 2.63 h (S.D. 1.42 h). There were no statistically significant differences between groups \((P > 0.05)\).

**Hypothesis testing: state and sex**

Skew of the distribution of each dependent variable was examined. Due to positive skew departures from normality for several measures, log 10 transformations were applied to cry latency, face latency, and cry duration, thereby improving the approximation to normal distributions.

**Facial action.** Individual facial actions of all types were summed across 2 segments (3 sec each) prior to heel-lance (during heel-rub) and separately for 2 segments (3 sec each) following heel-lance. A 3-way univariate ANOVA \((4 \times 2 \times 2)\) of summed facial movement across state, sex, and the heel-rub/heel-lance condition with repeated measures on the last factor was carried out.

Main effects of state, \( F(3, 131) = 7.29, P < 0.0001 \) and heel-rub/heel-lance, \( F(1, 132) = 295.35, P < 0.0001 \) were significant as was the state \( \times \) heel-rub/lance interaction, \( F(3, 132) = 3.69, P < 0.01 \). There were no significant sex differences.
The interaction was explored further with Duncan post-hoc comparisons separately across each level of the repeated measures factor with $\alpha = 0.05$ (see Table II).

Quiet awake (alert) infants responded with significantly more facial movement initially, as compared with those who were in quiet sleep, to both heel-rub and heel-lance stimulation. Additional state differences were apparent to a significant degree in heel-rub, namely quiet awake infants also showed more facial response than those in active sleep, and those in active awake more than those in quiet sleep.

For a more fine-grained assessment of different aspects of facial movement, log-linear analyses [22] of each facial action in each segment were carried out to examine the interrelationships of each face action with prior behavioral state and sex. Log-linear analyses showed only one statistically significant heel-rub effect for sex, namely brow bulge in segment 2, $\chi^2(3) = 8.28, P < 0.05$. No significant sleep/wake state by sex interactions were found. As 28 log-linear analyses were executed at a type I error rate of 0.05, this sole sex effect was considered a chance finding. With sex eliminated from consideration the log-linear analysis is reduced to a two-way $\chi^2$ test. Adjusted residuals were used to indicate which cells deviated from expected levels when overall significance was found. Frequency of taut tongue, $\chi^2(3) = 21.27, P < 0.0001$, vertical stretch mouth, $\chi^2(3) = 12.00, P < 0.007$, and naso-labial furrow, $\chi^2(3) = 10.81, P = 0.01$, showed statistically significant differences across state in segment 1. Similarly for segment 2, taut tongue, $\chi^2(3) = 13.55, P < 0.003$, vertical stretch mouth, $\chi^2(3) = 23.23, P < 0.001$, and naso-labial furrow, $\chi^2(3) = 14.99, P < 0.002$ varied by state. In segment 1 infants who had been quiet awake prior to heel-rub showing significantly more occurrences of each of these facial actions (40%, 25% and 65% respectively) than infants who had been in quiet sleep (6%, 4% and 24% respectively). Segment 2 showed the same pattern except for naso-labial furrow, where active awake and quiet awake were about equal. Eye squeeze differed significantly across state for segment 1, $\chi^2(3) = 8.76, P < 0.03$, and segment 2, $\chi^2(3) = 13.41, P < 0.004$. In segment 1 eye squeeze occurred most for quiet awake (65%) and least for quiet sleep (29%). Eye squeeze in segment 2 showed similar reactivity for both awake states (55% and 58%), and
significantly less for quiet sleep (20%). Lip part showed no statistically significant differences across sleep/waking states, but there appeared to be a trend toward less reaction by those who had been asleep with least occurrence following quiet sleep.

To heel-lance 96% of the infants showed brow action, 96% eye squeeze, 97% naso-labial furrow, and 98% lip part in segment 3. In segment 4, 96% of the quiet sleep infants responded with all 4 facial actions, and 100% of infants in the other 3 states. Thus, these face variables formed a constellation by virtue of their co-occurrence and were not significantly state dependent. In contrast, occurrence of stretch mouth and taut tongue were significantly related to sleep/waking state in segment 3, $\chi^2(3) = 7.65$, $P < 0.05$, and $\chi^2(3) = 8.54$, $P < 0.04$ respectively, but not in segment 4. Frequency of occurrence of taut tongue and stretch mouth following heel-lance was greatest for quiet awake infants (85% and 55% respectively) and least for quiet sleep (57% and 22%) respectively. The pattern of response occurrence to heel-rub (segments 1 and 2) and heel-lance (segments 3 and 4) across state for vertical stretch mouth and taut tongue is presented graphically in Figs. 1 and 2. Fig. 3 provides an artist’s rendering of the responses of two babies using video playback, one initially in quiet sleep, and the other quiet awake.
Latency to face movement. A 2-way 4 (state) × 2 (sex) ANOVA was carried out on the latency to face movement. There was a significant main effect for sex, $F(1, 132) = 4.73$, $P < 0.03$ (with shorter time to reaction for boys than girls), but not for state, and there was no significant interaction. Means and standard deviations across state and sex are shown in Table III.

Cry cycles, latency and duration of first cry to heel-lance. Number of cry cycles and duration of first cry to heel-lance were analyzed with univariate 2-way 4 (state) × 2 (sex) ANOVA. The means and standard deviations by sex and state are provided in Table III. Boys cried significantly more cycles than girls, $F(1, 132) = 3.74$, $P < 0.05$. The ANOVAs showed no significant differences across state and no sex by state interactions. As the distribution of cry latency remained significantly skewed despite the log 10 transformation, non-parametric 3-way log linear analysis was used [22]. The best fit model showed state, $\chi^2(3) = 8.18$, $P < 0.04$, and sex $\chi^2(1) = 5.93$, $P < 0.01$, as both statistically significant, but no significant interaction. Examination of ratios of the log-linear parameter estimates to standard error indicated boys cried sooner than girls, and infants in quiet sleep cried less quickly than the other states.
**Fundamental frequency.** Fundamental frequency of the first post heel-lance cry, measured at the first pitch-point ($f_1^1$), at 100 msec ($f_0^1$), and at 200 msec ($f_0^2$), was analyzed in a 3-way 4 (state) x 2 (sex) x 3 (time) ANOVA with repeated measures on the time factor. There were no main effects for state or sex and no significant interactions. The time factor was significant, $F(2, 234) = 14.26, P < 0.0001$, with a rising pattern evident over the 300 msec for boys and girls in each of the behavioral states.

**Pitch-plot pattern.** A pitch-plot was computer generated for the entire first cry to heel-lance for each infant. Each was classified into 1 of 6 categories based on the overall pattern. Approximately one-third of all the infants showed a rising-falling pitch pattern, regardless of the prior state. There were no apparent overall significant differences between the states.

**Interrelationships among facial actions.** It was of interest to examine whether different parts of the face were moving independently. Each facial action was summed across the 7 heel-rub and heel-lance segments combined, and Spearman

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**TABLE III**

**TEMPORAL CHARACTERISTICS OF THE RESPONSE TO HEEL-LANCE**

<table>
<thead>
<tr>
<th>Behavioral state</th>
<th>Quiet sleep</th>
<th>Active sleep</th>
<th>Quiet awake</th>
<th>Active awake</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Cry cycles</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Boys N</td>
<td>26</td>
<td>26</td>
<td>12</td>
<td>13</td>
</tr>
<tr>
<td>X</td>
<td>13.00</td>
<td>16.15</td>
<td>11.42</td>
<td>14.92</td>
</tr>
<tr>
<td>S.D.</td>
<td>6.11</td>
<td>5.44</td>
<td>5.37</td>
<td>6.8</td>
</tr>
<tr>
<td>Girls N</td>
<td>23</td>
<td>19</td>
<td>8</td>
<td>13</td>
</tr>
<tr>
<td>X</td>
<td>11.30</td>
<td>12.16</td>
<td>12.37</td>
<td>13.77</td>
</tr>
<tr>
<td>S.D.</td>
<td>5.58</td>
<td>5.87</td>
<td>4.41</td>
<td>7.46</td>
</tr>
<tr>
<td><strong>Cry duration</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Boys N</td>
<td>26</td>
<td>26</td>
<td>12</td>
<td>13</td>
</tr>
<tr>
<td>X</td>
<td>3.29</td>
<td>3.25</td>
<td>3.12</td>
<td>3.31</td>
</tr>
<tr>
<td>S.D.</td>
<td>0.45</td>
<td>0.51</td>
<td>0.55</td>
<td>0.41</td>
</tr>
<tr>
<td>Girls N</td>
<td>23</td>
<td>19</td>
<td>8</td>
<td>13</td>
</tr>
<tr>
<td>X</td>
<td>3.33</td>
<td>3.34</td>
<td>3.31</td>
<td>3.34</td>
</tr>
<tr>
<td>S.D.</td>
<td>0.43</td>
<td>0.42</td>
<td>0.62</td>
<td>0.45</td>
</tr>
<tr>
<td><strong>Latency to cry</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Boys N</td>
<td>26</td>
<td>26</td>
<td>12</td>
<td>13</td>
</tr>
<tr>
<td>X</td>
<td>2.26</td>
<td>1.49</td>
<td>2.29</td>
<td>2.29</td>
</tr>
<tr>
<td>S.D.</td>
<td>1.17</td>
<td>1.41</td>
<td>1.25</td>
<td>1.12</td>
</tr>
<tr>
<td>Girls N</td>
<td>23</td>
<td>19</td>
<td>8</td>
<td>13</td>
</tr>
<tr>
<td>X</td>
<td>2.77</td>
<td>2.44</td>
<td>1.97</td>
<td>2.06</td>
</tr>
<tr>
<td>S.D.</td>
<td>1.01</td>
<td>1.22</td>
<td>1.21</td>
<td>1.24</td>
</tr>
<tr>
<td><strong>Latency to facial movement</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Boys N</td>
<td>26</td>
<td>26</td>
<td>12</td>
<td>13</td>
</tr>
<tr>
<td>X</td>
<td>0.92</td>
<td>0.63</td>
<td>0.99</td>
<td>1.11</td>
</tr>
<tr>
<td>S.D.</td>
<td>0.54</td>
<td>0.56</td>
<td>1.07</td>
<td>0.94</td>
</tr>
<tr>
<td>Girls N</td>
<td>23</td>
<td>19</td>
<td>8</td>
<td>13</td>
</tr>
<tr>
<td>X</td>
<td>1.37</td>
<td>1.22</td>
<td>0.82</td>
<td>0.90</td>
</tr>
<tr>
<td>S.D.</td>
<td>0.99</td>
<td>0.74</td>
<td>0.56</td>
<td>0.69</td>
</tr>
</tbody>
</table>

---

*a* Log 10 msec.

*b* Log 10 sec.
TABLE IV
SPEARMAN CORRELATIONS BETWEEN FACIAL ACTIONS SUMMED OVER HEEL-RUB AND HEEL-LANCE SEGMENTS

<table>
<thead>
<tr>
<th></th>
<th>Eye squeeze</th>
<th>Naso-labial furrow</th>
<th>Lip part</th>
<th>Vertical stretch mouth</th>
<th>Taut tongue</th>
</tr>
</thead>
<tbody>
<tr>
<td>Brow</td>
<td>0.95 b</td>
<td>0.78 b</td>
<td>0.50 b</td>
<td>0.27 a</td>
<td>0.28 a</td>
</tr>
<tr>
<td>Eye squeeze</td>
<td>0.84 b</td>
<td>0.46 b</td>
<td>0.24 a</td>
<td>0.24 a</td>
<td></td>
</tr>
<tr>
<td>Naso-labial furrow</td>
<td>0.49 b</td>
<td></td>
<td>0.23 a</td>
<td>0.24 a</td>
<td></td>
</tr>
<tr>
<td>Lip part</td>
<td></td>
<td></td>
<td>0.14</td>
<td>0.23 a</td>
<td></td>
</tr>
<tr>
<td>Vertical stretch mouth</td>
<td></td>
<td></td>
<td></td>
<td>0.27 a</td>
<td></td>
</tr>
</tbody>
</table>

a P ≤ 0.01.
b P ≤ 0.001.

correlations were calculated (see Table IV). Eye squeeze, naso-labial furrow and brow action tended to occur simultaneously forming a cluster of facial actions. Mouth movements (lip part, vertical stretch and taut tongue) were significantly related to upper face action and naso-labial furrow, but only to a moderate degree. All infants showing vertical stretch mouth also had naso-labial furrow, but the correlation between the two was not high as there were many infants with naso-labial furrow whose mouths were open but not stretched. Lip part was the most common initial facial action to disturbance and, whereas all infants with taut tongue had their lips apart, the converse was not the case; thus there was no significant correlation. Of course, taut tongues could not be observed if lips were closed.

Discrete facial actions for segments 3–7 were summed to provide an overall measure of facial movement to heel-lance. Latency to facial movement was significantly negatively correlated with total facial action (Pearson $r = -0.52$, P < 0.001). Thus, faster initial face movement was associated with greater facial activity.

Interrelationships among cry measures. Pearson correlations among the cry measures were calculated. The number of cry cycles following heel-lance was unrelated to either the speed of cry reaction or to the duration of the first cry. The only statistically significant correlation at P ≤ 0.01 was between fundamental frequency at 300 msec and speed of cry response but it was of small magnitude ($r = 0.26$).

TABLE V
PEARSON CORRELATIONS BETWEEN CRY MEASURES AND FACE ACTIONS TO HEEL-LANCE

<table>
<thead>
<tr>
<th></th>
<th>$t_0^1$</th>
<th>$t_0^2$</th>
<th>$t_0^3$</th>
<th>Number of cry cycles</th>
<th>Duration of 1st cry</th>
<th>Latency to cry</th>
</tr>
</thead>
<tbody>
<tr>
<td>Latency to</td>
<td>0.00</td>
<td>0.15</td>
<td>0.13</td>
<td>-0.07</td>
<td>0.13</td>
<td>0.53 b</td>
</tr>
<tr>
<td>facial movement</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total face</td>
<td>0.10</td>
<td>0.03</td>
<td>0.01</td>
<td>0.24 a</td>
<td>0.00</td>
<td>-0.34 b</td>
</tr>
<tr>
<td>action</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

a P ≤ 0.01.
b P ≤ 0.001.
Relations between cry measures and face movement. As shown in Table V, speed of facial movement to heel-lance was significantly correlated with speed of cry response. Total facial action following heel-lance was negatively correlated with latency to cry, indicating faster speed to cry was related to amount of face action. Also, the number of cry cycles following heel-lance was significantly correlated with amount of facial action, but only to a rather moderate degree ($r = 0.24$). This suggested that many babies who cried only a few cycles moved their faces a good deal. Fundamental frequency showed no relation to facial movement.

Discussion

Distinguishing between ‘pain’ as it is commonly construed from the adult perspective [10] and ‘reflexive’ responding has been an issue when considering the nature of the newborn’s response to invasive procedures [4,34]. Pain remains a construct attempting to account for subjective experiences and is inferred from observation of tissue damage and behavior. Following heel-lance, neonates early in the second day of life showed a constellation of facial changes, namely eye squeeze, brow contraction, naso-labial furrow, taut tongue and open mouth, together with cry response, that differed substantially from the amount and type of facial action provoked by heel-rub. This reaction pattern may be described operationally as ‘pain’ expression. Doubtless the reaction pattern observed is a complex reflexive response. In bioevolutionary terms, for species survival neonatal sensitivity to pain and a highly developed means of communicating pain to the caretaker appear essential and fundamental. In turn, adults are likely to interpret the behavioral display as indicative of at least the rudimentary qualities of pain experience [32]. Indeed, feelings of concern for the babies in pain represent one’s strong reaction when viewing these babies.

Infant studies in the last two decades have shown a far greater degree of competence and behavioral organization in infants and newborns than previously believed [9,40]. The present study demonstrates a cohesive response pattern to invasive stimulation, which is consistent with the response capacities currently recognized in the other major sensory modalities. But, the reaction pattern was not exclusively the imperative response to the tissue insult. Pain expression in newborns was found to be a function of ongoing behavioral state, rather than solely reflecting tissue damage. Due to the limited opportunity and capacity for neonates to have learned secondary responses to nociceptive input (although intrauterine learning remains a possibility), the differences in facial expression and latency to facial movement and to cry across state were interpreted as reflecting differences in the inherent capacity for modulation of pain perception.

Of the alternatives hypothesized to understand how infant responsivity could vary across state, Brazelton’s view [7,8] of the alert awake state as providing optimal orientation or receptivity to environmental input received the strongest support.

The facial actions examined here which persisted in significantly differentiating responses between states were both oral, namely, vertical stretch mouth and taut
tongue. Oral functioning has been viewed as the highest level of coordinated functioning of the newborn due to the fact that it is so intrinsic to survival [40]. The significance of non-nutritive oral behavior is unclear, however it has been regarded as a sensitive indicator of arousal [25].

The results did not support Levine and Gordon's [27] proposal that pain induced vocalization would be a sensitive indicator of infant pain perception, insofar as pain experience differed across sleep/waking states. Levine and Gordon's position was mainly derived from studies of vocalization in infrahuman species, in which distinct 'calls' have been identified. Fundamental frequency of pain cry has been found to be related to the health status of neonates, and has been viewed as indicative of recovery capacity following stimulation [26]. It was expected that where more facial activity was evident, fundamental frequency would vary accordingly, but the data did not support this perspective. The results suggested that the view from studies of 'at risk' infants, suggesting fundamental frequency reflects capacity for response modulation, does not generalize to healthy infants across sleep/waking states. It appears that whereas unwell infants may have difficulty modulating their responses to stimulation, and this may be reflected in higher fundamental frequency of initial pain cry, healthy neonates have excellent adaptive capacities as part of their biological survival mechanisms.

Sex differences were apparent, but only in the domains of response latency and number of cry cycles, which supports Brackbill's and Shroder's [6] position that there is not strong evidence for behavioral differences between male and female neonates. However, our findings of shorter speed of response for males were consistent with observations of response to pin-prick during administration of the Brazelton Neonatal Behavioral Scale [7]. Sex differences which may exist in response to noxious insult appear to depend on parameters not yet delineated or understood.

One of the main contributions of this study was in the area of neonatal pain measurement. As with adults [12], discrete facial action appeared to be a reliable and useful measure of pain expression. Although the infants showed a consistent reaction pattern to pain, the variability in this display pattern across states was informative. Two aspects were isolated as particularly sensitive to state variation and worthy of further study in their own right, namely taut tongue and vertical stretch mouth. The facial measures varied systematically with types of intervention (heel-rub and heel-lance). There is evidence that neonatal facial activity systematically varies as a function of obstetric events during childbirth [19]. It was noteworthy that the use of a coding system which did not limit the investigator to a preselected configuration of facial activity as depicting emotional categories or pain actually promoted the discovery of 'new' information, as any visually detectable category of movement on the video frame qualified for possible attention as a 'facial variable.'

Highly significant differences in infant facial activity during the latter phase of blood collection as a function of laboratory technician were observed. Although this portion of the procedure was not used in the data analyses, it was of interest in itself as an indication of neonates surprising sensitivity to variations in handling. In the
light of the prevalent interest in the importance of caretaker–infant transactional behavior, and contingent responding to infant cues [8,41], interaction patterns during medical procedures certainly warrant systematic study. As ongoing functional state modified pain behavior, further study of pain expression using the proposition that ongoing events alter pain perception may be fruitful. Within this model it would be expected that other factors either intrinsic to the individual neonate such as temperamental variation, or extrinsic variables such as soothing or swaddling during the stimulation, may also affect pain behavior.

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