Dissociation of heart rate and somatomotor orienting responses to novel stimuli in preweanling rats

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Changes in heart rate (HR) and somatomotor behavior elicited by the presentation of novel olfactory, visual, or auditory stimuli were examined in preweanling rat pups, 20-21 days of age. Pups exposed to an amyl acetate odor, a pulsating tone, or a flashing light showed the marked cardiac deceleration characteristic of the orienting response. In contrast, the three stimuli elicited distinct somatomotor behavioral patterns. Pups exposed to a novel amyl acetate odor or pulsating light raised their heads and reared, whereas animals exposed to a novel pulsating tone (16 kHz, 85 dB) performed a lateral head jerk and then maintained a relatively fixed posture. Both HR and gross motor activity to the novel stimulus displayed rapid habituation. This report provides clear evidence for a dissociation between somatomotor activity and HR as components of the orienting response in the developing rat.

Heart rate (HR) has proved to be a useful tool in the analysis of both stimulus receptivity (e.g., Campbell & Haroutunian, 1983; Graham & Clifton, 1966) and basic learning mechanisms (Campbell & Ampuero, 1985; Martin & Fitzgerald, 1980; Fitzgerald, Stainbrook, & Francisco, 1985). However, one concern facing investigators who utilize HR as an index of learning or stimulus orienting is the possibility that HR may be an artifact of somatomotor activation or inhibition. Cardiosomatic coupling refers to the possibility that HR covaries with levels of somatomotor activation (Obrist, 1981).

When an organism is presented with an innocuous novel stimulus, HR decreases. This deceleration in HR is commonly known as the heart rate orienting response (HR OR) and is distinguished from the acceleratory defensive response. The former is thought to enhance stimulus processing, whereas the latter reduces or limits it (Graham & Clifton, 1966; B. Lacey & J. I. Lacey, 1964; J. I. Lacey & B. C. Lacey, 1958; Sokolov, 1963). Most researchers interpret the HR OR to be a reflection of stimulus receptivity. It is possible, however, that cardiosomatic coupling may account for this phenomenon. Perhaps, when an animal first encounters a novel stimulus, it freezes, and this freezing response causes HR to decrease. Partial support for cardiosomatic coupling in stimulus orienting has been obtained in human infants. Pomerleau-Malcuit, Malcuit, and Clifton (1975) demonstrated that

cheek stroking elicited cardiac deceleration when no overt behavioral response was observed, but that when the tactile stimulation elicited head turning, it was accompanied by cardiac acceleration. However, Pomerleau-Malcuit et al. also found that an ear pinch, which produced no movement, led to greater tachycardia than cheek stroking that elicited movement. This pattern of interactions argues against any simple cardiosomatic explanation of their results.

These findings are in agreement with the animal literature. Upon presentation of a novel stimulus, restrained adult rats placed on a movement-sensitive surface display cardiac deceleration while exhibiting somatomotor activation (Fitzgerald et al., 1985). More recently, using frequency of interruptions of the electrocardiogram (EKG) as their index of somatomotor activation, Dailey, Wigal, and Amsel (1986) found no relationship between HR and somatic activity when young rats oriented to a photic stimulus.

The purpose of the following experiments was to further examine the relationship between cardiac deceleration and somatomotor activation during orienting in preweanling rats. Our approach was to systematically observe, while simultaneously recording HR, the behavioral responses of freely moving young rats when they were presented with a novel stimulus. In three separate experiments, subjects were exposed to a novel odor, a flashing light, or a pulsating tone.

GENERAL METHODS

Subjects

The subjects were 24 experimentally naive male and female rat pups derived from the Sprague-Dawley strain bred and reared in the animal colony of the Psychology Department at Princeton University. The day of birth was counted as Day 0. All litters were

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culled to 8 pups at 2 days of age and were left undisturbed until the day of experimentation. The pups were housed in 45×24×21 cm plastic rat cages with the dam, in a colony maintained on a 16/8 day/night cycle at 23°±1°C. Purina Rat Chow and water were available ad lib. The subjects were 20–21 days of age at the time of testing. No more than 2 animals from any one litter were used in testing.

HR Recording Procedure

Heart rate was recorded through two subcutaneous stainless steel electrodes in the form of safety pins attached to 32-ga Teflon-covered stainless steel wire (Alpha 2840/7). One was implanted dorsally, at the nape of the neck, and the other was implanted ventrally. Neither the HR electrodes nor the attached leads appeared to interfere with the pups' movement. The cardiac potential thus obtained was amplified by a Grass Model P15 amplifier. The R spike activated a Coulbourn Instruments Schmitt trigger. This pulse was fed to a Commodore PET microprocessor that measured the interbeat interval (IBI) to the nearest millisecond and stored that information on floppy diskettes for further analysis. The microprocessor also controlled stimulus presentation and all timing sequences of the experiment. The cardiac signal was continuously displayed on a Hitachi oscilloscope (Model V-212).

The animals were removed from the home cage and brought to the test chamber, where the electrodes were inserted. This procedure typically took 10–20 sec. All animals were then placed in the test chamber for a 15-min adaptation period prior to the stimulus presentation.

Behavioral Observation

Somatomotor behaviors were scored on the basis of preliminary observations. Behaviors scored were: immobile, head jerk, raise head, and walk/rotate. Table 1 lists the criteria for behavioral scoring.

The behavior of the rats was scored by a single observer during the 5-sec prestimulus baseline sample and the 10-sec stimulus presentation. During randomly selected sessions, a second observer was also present. In each of these sessions there was an agreement of over 90% between observers. If more than two types of behavior were observed during a 5-sec period, only the two dominant patterns were scored. Scores for each 5-sec period for Trials 1, 5, and 10 were tallied and divided by the number of animals, to give the percentage of animals displaying the particular behavior.

EXPERIMENT 1: ODOR

Method

Subjects. Eight experimentally naive male and female preweanling rats, born and bred in the manner previously described, were used in this experiment.

Apparatus. An amyl acetate odor stimulus was presented to the pups in a Plexiglas cage (28×20×60 cm). The cage had a stainless steel grid floor and was housed in a sound-attenuating chamber that was illuminated by two 6-W shielded nightlights placed in the rear corners of the chamber. A gentle stream of warm air, the main airflow, passed through the test chamber continuously throughout the test session. Air entered the Plexiglas chamber from the bottom and exited at the top. It was delivered by an airpump (Thomas 607C/EF32) located outside the test room, and was exhausted at the other end of the chamber by negative pressure generated by an exhaust fan. This airstream was kept at a constant temperature (27°C) by being passed through a heated, temperature-regulated water coil. Another, smaller airstream, generated from the same airpump, passed through either a flask containing 40 ml of distilled water, to produce a "neutral airstream," or a flask containing a .1% amyl acetate solution (100 ml), to produce

Table 1
Criteria for Behavioral Scoring

Immobile:	Animal is stationary. No overt movement observed.
Raise Head:	Lifting of the head in gradual fashion. Often preceded by sniffing and/or slow movement of the head from side to side. (Rearing frequently followed head raising, but was not scored separately since it seemed to be a simple continuation of the initial sequence of behavior.)
Head Jerk:	Short, ballistic lateral movement of head. Animal remains motionless except for movement of the head.
Walk/Rotate:	Any walking or circling. Frequently accompanied by sniffing.

an "odorized airstream." This airstream, which was controlled by two "T" valves, one before the two flasks and one after, joined the main air flow shortly before entering the Plexiglas test chamber. The neutral airstream remained on throughout the session except during the 10-sec presentation of the conditioned stimulus (CS). During this period, the valves were switched, shunting the small airstream from the water flask to the odorant flask. The odor was evacuated along with the main airflow at the other end of the test chamber by negative pressure generated by the exhaust fan. To mask the brief clicking sound of the opening and closing of the mechanical valves, random clicking sounds were generated throughout the session by the activation of an additional valve, mounted adjacent to the main valve.

Procedure. All subjects received 10 presentations of the amyl acetate odor stimulus on a variable 180-sec intertrial interval (range 75–285 sec). The four previously described behaviors were scored once during the 5-sec prestimulus period and twice during the 10-sec stimulus presentation.

Results

The data for the animals exposed to the amyl acetate odor are displayed in Figure 1. Data are presented for Trials 1, 5, and 10 for the 5-sec prestimulus baseline period (pre) and the 10-sec stimulus period, divided into two 5-sec bins (5 and 10). The top panel displays HR in beats per minute (BPM). The next four panels show the percentages of animals that remained immobile, raised their heads, displayed the head-jerk reaction, and walked or rotated (second through fifth panels, respectively).

Upon presentation of the novel odor (Trial 1), HR decelerated while animals exhibited behavioral activation. The behavioral activation is seen in the decrease in the percentage of animals that remained immobile during stimulus presentation. A within-subjects *t* test (Runyon & Haber, 1980) showed the cardiac deceleration to be significant [$t(7) = 3.70, p < .01$], and the Cochran *Q* test (Siegel, 1956) revealed the decrease in the percentage of animals that remained immobile during stimulus presentations to be significant ($Q = 11.1, p < .01$). The behavioral reaction seen in response to the novel olfactory stimulus was quite striking. Upon stimulus onset, animals would begin to vigorously sniff the air, and, while moving their heads from side to side in slow sweeping motions, they would gradually lift their heads. Rearing frequently accompanied these behaviors. During the 5-sec prestimulus baseline measure, none of the animals raised

Figure 1. Cardiac (top panel) and behavioral (bottom four panels) responses to an amyl acetate odor during a 5-sec prestimulus period and a 10-sec stimulus period.

their heads, but during the first 5 sec of the stimulus presentation, 63% of the animals showed this behavior, and during the second 5 sec, 82% displayed it ($Q = 11.1$, $p < .01$). By Trial 5, both HR and behaviors had habituated to the odor (all $ps > .05$).

These observations and recordings demonstrate that presentation of an unfamiliar odor, amyl acetate, simultaneously produces a decrease in HR and an increase in behavioral activation. No simple models of cardiosomatic coupling account for these findings.

EXPERIMENT 2: LIGHT

Method

Subjects. Eight experimentally naive preweanling pups (21 days old) of both sexes, born and bred as previously described, were used in this experiment.

Apparatus. The behavioral apparatus consisted of a 22×22×50 cm topless Plexiglas cage located inside a 128×77×48 cm sound- and light-attenuating chamber (IAC), maintained at ambient temperature ($23^{\circ} \pm 1^{\circ} \text{C}$). A 60-W light bulb was suspended outside and to the right of the Plexiglas cage, 18 cm from the floor of the outer chamber. The light stimulus was flashed on and off at a rate of 250 msec on and 250 msec off. Dim red illumination, supplied by

a single 6-W nightlight placed in a cone of red cellophane located in the corner of the apparatus, provided sufficient illumination for observation of the animals' behavior during the prestimulus period.

Procedure. All subjects were given 10 presentations of the flashing-light stimulus on a variable 180-sec intertrial interval (range 75–285 sec). As in the previous experiment, the behavior of the animals was observed and scored during the 5-sec prestimulus and 10-sec stimulus periods.

Results

Figure 2 displays the data for the animals exposed to the pulsating light and is organized in the same fashion as Figure 1. The top panel shows that upon presentation of the flashing light (Trial 1), HR decreases [$t(7) = 8.00$, $p < .01$] while the animals again displayed behavioral activation. This is evident in the decrease in the percentage of animals remaining immobile during presentation of the flashing light (Panel 2). As the third panel for Trial 1 shows, behavioral activation is primarily the result of the animals' raising their heads in response to the stimulus ($Q = 8.11$, $p < .05$).

The behaviors observed when animals oriented to the flashing light were quite similar to those obtained with novel odor, except for the absence of vigorous sniffing

Figure 2. Cardiac (top panel) and behavioral (bottom four panels) responses to a pulsating 60-W light during a 5-sec prestimulus and a 10-sec stimulus period.

and profound sweeping motions of the head. The animals did, however, occasionally sniff the air and move their heads from side to side while gradually lifting their heads. By Trial 5, both HR and the observed behaviors had habituated (all $ps > .05$).

As in the previous experiment, when the animals were presented with a novel stimulus, HR decreased while subjects exhibited behavioral activation. Again, as with an unfamiliar odor, cardiosomatic coupling cannot account for the decrease in HR seen when animals orient to a novel visual stimulus.

EXPERIMENT 3: TONE

Method

Subjects. Eight experimentally naive male and female preweanling rats, born and bred in the manner previously described, were used in this experiment.

Apparatus. The chamber was the one used in Experiment 2. Dim illumination in the IAC was supplied by two 6-W shielded night-lights placed in the far corners of the chamber. An 85-dB, 1600-Hz tone, generated by a Coulbourn Instruments precision signal generator (Model S81-06), attenuated by a Hewlett-Packard attenu-

ator (Model 350-D), amplified by a Realistic solid state stereo amplifier (Model Sa-10), and delivered through an 8-in. speaker mounted on the ceiling of the sound-attenuating chamber, was used at a rate of 250 msec on/250 msec off.

Procedure. All subjects were given 10 presentations of the tone stimulus on a 180-sec intertrial interval (range 75–285 sec).

Results

The data for animals exposed to the pulsating tone are presented in Figure 3; they are organized in the same manner as in the previous figures. Upon presentation of the novel auditory stimulus, HR decreased significantly from baseline levels [$t(7) = 3.71, p < .01$] while the animals simultaneously displayed behavioral activation ($Q = 6.33, p < .05$). This behavioral activation was primarily in the form of a head-jerk response (Panel 4) rather than head raising as with the previous stimuli (Panel 3). The head jerk consisted of an abrupt, ballistic, lateral motion of the head. The animals typically performed one head jerk and then maintained their posture throughout the rest of the stimulus period. This accounts for the decrease in the percentage of animals showing the head-jerk response during the last 5 sec of the stimulus period (37%) as com-

Figure 3. Cardiac (top panel) and behavioral (bottom four panels) responses to a 1600-Hz (85-dB) pulsating tone during a 5-sec prestimulus and a 10-sec stimulus period.

pared with the first 5 sec (63%). By Trial 5, both HR and behaviors had habituated to the pulsating tone (all $ps > .05$).

With the auditory stimulus, the primary somatomotor reaction, a ballistic movement of the head, was much shorter in duration than the head-raising and sniffing responses elicited by olfactory and visual stimuli. Although the tone cannot be described as inhibiting movement, since the animals were quiescent prior to stimulus onset, the overall pattern of behavior (i.e., maintenance of posture) is compatible with the view that HR deceleration is associated with inhibition of somatomotor activity. However, when the results from the three experiments are considered, it is clear that the pattern of somatomotor activity elicited by brief sensory stimuli does not determine either the direction or magnitude of the HR response.

GENERAL DISCUSSION

These experiments demonstrate that novel stimuli in different sensory modalities produce a decrease in HR, typical of the OR, and a characteristic, modality-specific pattern of increased somatomotor activation. The results

obtained suggest that the bradycardia seen during orienting is reflective of stimulus receptivity rather than cardio-somatic coupling.

The data corroborate previous reports which indirectly indicate that the deceleratory HR OR is not linked to somatomotor freezing. As their index of somatomotor activation, Fitzgerald et al. (1985) used the perturbation of a movement-sensitive surface by restrained rats, and Dailey et al. (1986) used the frequency of EKG interruption due to excessive movement. Both found that cardiac and somatic activity were not related during orienting. We, on the other hand, directly observed the behavioral responses of freely moving young rats to a novel stimulus while simultaneously recording heart rate. It is quite evident that animals do not freeze or inhibit activity when presented with a nonthreatening novel stimulus. Furthermore, our finding of modality-specific behavioral orienting is in agreement with the previous finding that adult rats display differential behaviors when exposed to a novel light or tone (Holland, 1977). Both behavioral and cardiac ORs exhibited rapid habituation to repeated stimulus presentation. The fact that behaviors were variable while HR was consistently deceleratory across all three sensory mo-

dalities provides further evidence for the usefulness of HR as a relatively stable index of stimulus orienting.

Animals presented with a novel odor or light raise their heads and rear while exhibiting bradycardia. These findings are not in agreement with those of Vanderwolf and Vanderwart (1970), who observed increased levels of HR during spontaneous rearing. Procedural differences may account for this discrepancy. Vanderwolf and Vanderwart measured basal levels of HR in adult animals over a 30-min period, and we measured the acute response of young animals to a novel stimulus. Our findings show unequivocally that HR can decrease even when animals go from a state of behavioral quiescence to behavioral activation. Thus, no simple correlation between cardiac and somatomotor activity can account for the decrease in HR seen during stimulus orienting.

There appears to be a similar lack of correlation between somatomotor activation and classical conditioning of HR. Numerous studies have shown that HR-conditioned responses can occur in the absence of any movement (Bruner, 1969; Cohen, 1969; Fitzgerald, 1966; Martin & Fitzgerald, 1980; Smith & Stebbins, 1965). Furthermore, Fitzgerald et al. (1985) observed conditioned cardiac deceleration in animals exhibiting somatomotor activation. Similarly, recent work in our laboratory on the developing rat has shown equivalent HR conditioning to an olfactory CS in both active and inactive infant animals (Sananes, Gaddy, & Campbell, in press).

Although cardiosomatic coupling does not account for the deceleratory HR OR, this does not rule out the possibility that HR may be linked to somatomotor activity in other situations. For example, it has been reported that basal levels of HR are higher in active than in inactive animals (Black, Fowler, & Kimbrell, 1964; Vanderwolf & Vanderwart, 1970). The correlation between higher HRs and spontaneous activity in the rat does not become striking until late in the preweanling period (Hofer & Reiser, 1969).

It is noteworthy that the behaviors observed were quite appropriate to the respective stimuli. An animal exposed to a novel odor vigorously sniffs the air, raises its head, and rears, whereas an animal exposed to a novel auditory stimulus jerks its head and maintains posture, apparently trying to localize sounds in space. An animal exposed to a novel pulsating light behaves much the same as an animal that encounters a novel odor; when presented with the novel visual stimulus, the animals also sniff the air, raise their heads, and rear. The fact that young albino rats have poor eyesight may account for their reliance on other sensory modalities (e.g., olfaction) to aid in discerning novel visual stimuli encountered.

To conclude, the important outcome of this research is the finding that the cardiac deceleration component of the OR is not an artifact of somatomotor inhibition. We interpret the phenomenon of the HR OR to be reflective of stimulus receptivity and not cardiosomatic coupling. The finding that the HR component of the OR is independent of the somatomotor reaction increases its potential

usefulness as an index of sensory information processing in the developing as well as the adult animal.

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