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Cortisol, prolactin, growth hormone and neurovegetative responses to emotions elicited during an hypnoidal state

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Abstract

The present study describes the responses of cortisol, prolactin and growth hormone (GH) to emotions elicited during sessions in which an hypnoidal state was induced. The purpose of the study was to provide answers for the following questions: 1) Do sessions with an emotional content have more hormonal surges than baseline, relaxation-only, sessions? 2) Does the induction of a fantasy of pregnancy and nursing elicit a prolactin response? 3) Are there any associations between surges of different hormones? 4) Are hormonal responses related to the intensity, type, or mode of expression of the emotions? For this purpose, thirteen volunteers and twelve patients with minor emotional difficulties were studied during sessions under hypnosis. The period of observation lasted for about three hours. Heart rate (HR), skin conductance (SC) and vagal tone (VT) were monitored. Serum cortisol, prolactin and growth hormone were sampled every 15 minutes. The volunteers had three types of sessions— "blank", consisting of relaxation only (12 sessions), "breast feeding", in which a fantasy of pregnancy and breast feeding was induced (12 sessions) and "free associations" in which the subjects were encouraged to evoke experiences or feelings (17 sessions). The patients had only sessions of free associations (38 sessions). Sessions of free associations had more hormonal surges than "blank" and "breast feeding" sessions. This was true for cortisol (8/17 v.3/24; $p < 0.03$), prolactin (7/17 v. 3/24; $p < 0.05$) and GH (9/17 v. 4/24; $p < 0.02$). During the 55 sessions

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of free associations (volunteers plus patients) there were 32 surges of cortisol, 18 of prolactin and 28 of GH. Cortisol and prolactin surges were negatively correlated ($p < 0.03$). GH had no significant association with either cortisol or prolactin. Visible emotions were positively associated with GH surges ($p < 0.05$). but not with cortisol or prolactin. Cortisol surges were correlated positively with evocations of real events ($p < 0.01$) and negatively with evocations containing defensive elements ($p < 0.01$). Cortisol correlated positively with shock and intimidation ($p < 0.02$) and negatively with rage ($p < 0.04$). The AUC of the cortisol peaks during shock and intimidation was significantly higher than that of the pool of all other cortisol peaks (12.4 µmol.min.l^{-1} v. 7.1 µmol.min.l^{-1}; $p < 0.005$). Rage had a marginally significant positive association with prolactin surges (p=0.07). The distribution of GH surges did not show any significant association with types of emotions.

The present study provides evidence that cortisol, prolactin and GH respond to psychological stress in humans. However, they are regulated differently from one another. Cortisol and prolactin surges appear to be alternative forms of response to specific emotions. GH surges depend on the intensity of the emotion, probably as a consequence of the associated muscular activity. The current paradigm of stress, implying corticotrophin-releasing hormone (CRH) as the initial step of a cascade of events, is insufficient to account for the diversity of hormonal changes observed in psychological stress in humans.

Keywords: Cortisol; Growth hormone; Hypnosis; Prolactin; Psychological stress; Vagal tone

1. Introduction

Cortisol, prolactin and GH surges have been inconstantly observed following psychological stress in humans. However, it is unclear whether they are part of a generalized hormonal response, are steps of a staggered response or are independent responses to specific stimuli. In most experimental protocols hormonal plasma concentrations are measured in subjects exposed to typified stressors or during typified real life situations. Other quantities like perceived intensity of the stress, personality, coping styles and social support have been, occasionally, assessed. A comprehensive review of the reports published over the last two decades on neuroendocrine responses to psychological stress in humans appeared recently (Biondi and Picardi, 1999).

In the well described "stress" paradigm of physical aggression, the sympatho-adrenomedullary system and the hypothalamo-pituitary-adrenal (HPA) axis play central roles integrated by corticotrophin releasing hormone (CRH) (Fisher and Brown, 1991; Sapolsky et al., 2000).

Prolactin and GH are inhibited, rather than stimulated, by CRH, at least in rats (Rivier & Vale, 1985; Almeida et al., 1994). Surges in the plasma concentrations of these hormones have, occasionally, been observed during psychological stress in humans (Biondi and Picardi, 1999). Yet, they have been given little place in the theory of stress and no adaptive value has been attributed to them.

The parallelism between physical and psychological stresses should be viewed with caution. Activation of the HPA axis in physical stress is driven by inflammation or gross metabolic derangement (hypovolemia, hypoglycemia, etc.). The adaptive value of the HPA activation, apart from improving haemodynamics and fuel availability, consists of a negative feed-back effect to the inflammatory and vasoconstrictor responses (Sapolsky et al., 2000).

The adaptive value of increased corticosteroid levels during psychological stress probably depends on their actions on neural centers, including the hippocampus, that affect the processes of learning (Roozendaal, 2000).

Different from cortisol, prolactin activates maternal behavior, fat accumulation and the production of milk—all adaptive for the care of the young. Elevated prolactin values or galactorrhea have been associated with psychological stress and with situations related to the assumption of a parental role such as pseudopregnancy or the nursing of adopted children. The role of prolactin as part of a neuroendocrine subroutine adaptive to parenthood but susceptible to be associated with psychosomatic disorders has been recently reviewed (Sobrinho, 2001).

Some of us (M.S. and L.B.) have been actively involved in the development and usage of hypnosis for therapeutic purposes. Therefore, we decided to investigate the responses of prolactin, cortisol and GH and of the autonomic system—skin conductance (SC), heart rate (HR) and vagal tone (VT)—during sessions in which such states were induced.

The present study was designed primarily to answer the following questions: 1) Are there more hormonal surges during sessions in which emotions are evoked as compared to sessions of relaxation only?; 2) Does induction of a state of imagined breast feeding elicit a prolactin surge?; 3) Is there any association between surges of cortisol, prolactin and GH? 4) Is there any relationship between the intensity, type, or mode of expression of the evoked emotions and hormonal surges?

2. Patients and methods

2.1. Patients

First study—Thirteen healthy females aged between 21 and 53 were studied. They were recruited from a universe of medical students, psychologists or candidates to psychotherapy who volunteered for the study after its purposes and protocol were extensively discussed with them. The protocol was approved by the Ethics Committee of the Portuguese Cancer Institute. None of the subjects was taking any medication, except hormonal contraceptives, at the time of the study.

Second study—Twelve patients (eleven females and one male) recruited from the same universe as above, agreed to have their therapeutic sessions monitored. These patients were to initiate interpersonal psychotherapy for anxiety, minor depressive states or phobias. Three of the patients met DSM IV criteria for specific phobias—to needles, to glass and of being touched on the navel. One the subjects in the first study also revealed, during the preliminary interview, to have a specific phobia to the sea. None of the other subjects of either study met DSM IV criteria for any specific psychiatric condition.

All subjects were Caucasian. All, but patient 3 from study 2 were females. Other relevant characteristics of the populations studied are summarized in Table 1.

2.2. Protocol

The studies were carried out in the Department of Endocrinology of the Portuguese Cancer Institute.

By 1000 h the subjects were put comfortably in a quiet room, with natural light but in semi-obscurity. An indwelling Butterfly needle was placed in a forearm vein and electrodes for the recording of SC and electrocardiogram (ECG) were applied. The subject rested for about 60 min (stage 1) talking freely with one of the members of the team. During this period the therapist (MS or LB) prepared the patient for the experience to follow during the hypnoidal state. In the case of the sessions of free associations patient and therapist agreed on a theme to be addressed during the session. Subsequently, the psychotherapist induced a state of relaxation (stage 2)

Table 1
Characteristics of the populations studied

Subject	Group	Age	School	Marital status	Par	OC
1	1	53	S	M	1	0
2	1	21	U	S	0	0
3	1	22	U	S	0	0
4	1	39	U	D	1	0
5	1	31	S	M	0	0
6	1	32	U	M	1	0
7	1	21	C	S	0	1
8	1	30	U	M	1	0
9	1	21	U	S	0	0
10	1	45	U	M	1	0
11	1	34	U	M	1	0
12	1	22	C	S	0	1
13	1	23	U	S	0	0
1	2	46	U	D	1	0
2	2	31	C	M	0	0
3	2	36	U	M	—	—
4	2	21	U	s	0	0
5	2	22	U	s	0	0
6	2	23	U	M	1	0
7	2	19	U	S	0	1
8	2	22	U	S	0	0
9	2	54	C	M	1	0
10	2	21	U	S	0	1
11	2	38	U	M	1	0
12	2	25	U	S	0	0

Subj: Subject number; School: Scholarity; U: University; C: College; S: Secondary studies; Marital Status; M: Married; S: Single; D: Divorced; Par: Parity; 1: Parous; 0: Nulliparous; OC: Oral contraceptives; 1: Yes; 0: No.

according to Kroger (1960). A deepening of this state was obtained through the visualization of a peaceful landscape so that the patient was kept in a state of waking hypnosis (Kroger, 1977: pp. 81–84). The induction of this state (stage 2) took about twenty minutes. In this state the subjects are deeply relaxed but conscious, keeping an interactive contact with the therapist and verbalizing what they are experiencing. Under deep relaxation the subjects are told to experience an imagined situation or, in the case of free associations, to freely associate with a feeling or actual scene of their past, related to the theme previously selected. Each evocation can last from a few minutes to the entire session. When an episode appears to be drained out, the therapist encourages the patient to associate with another episode related to the initial theme. The patients enter a state of self induced "virtual reality" in which the experienced scene is cathartic. Towards the end of the session, an attempt is made to use the obtained insights to provoke changes in the cognitive behavior pattern (Simões et al., 1999).

The hypnoidal state lasted one hour and was followed by a second period of relaxation before awaking (stage 4) of about 10 min. Finally, after the patients were fully awake, they were debriefed for about 15 min (stage 5). The contents, feelings and emotions expressed during the sessions were then discussed, further associations were made, the cognitive redefinition of the expressed material was reinforced and plans were made for follow up.

The whole process lasted about three hours.

2.2.1. First study

Each subject had three sessions of different types, always in the same order: 1—A "blank" session in which the subjects dozed most of the time. The therapist made occasional interventions to prevent them from falling asleep. The actual situation to be induced was negotiated during the preparatory stage of the session to avoid possible associations with traumatic events. 2—A "breast feeding" session during which a fantasy of breast feeding was induced, starting from the desire to have a child, the fulfillment of the expectations of pregnancy, the relation with the child during the course of the pregnancy, giving birth, nursing and the dialogues with the child during nursing; 3—A session consisting of free associations about a feeling or event previously selected by the subject. One of the subjects had three sessions of free associations. One subject, who was actually nursing a 4 month old child, had three "breast feeding" sessions only.

2.2.2. Second study

Each patient had 1 to 8 sessions identical in design to the free associations sessions of the first study. The total number of these sessions was 38. For the purposes of the calculations, the 17 sessions of free associations from the first study were added to this group. This merging of the observations was considered to be legitimate because all the patients had been recruited from the same universe, none of them had any major psychiatric disorder and the intensity, mode of expression and type of the emotions displayed was similar for both groups of subjects, as were the demographic characteristics (Table 1), the neurovegetative and the hormonal responses (see Results).

2.3. Methods

2.3.1. Recording of SC and ECG

SC was measured with a 2701 Bioderm SC Monitor (UFI, CA, USA). The signal was recorded every half second and was expressed in μS. HR and VT were extracted by a VT Monitor (Delta-Biometrics Inc., Bethesda, MD, USA), as follows. The record of the R-R intervals was edited with the MXedit software, also from Delta-Biometrics. VT estimates were taken as natural logarithm of the variance of the R-R intervals in the frequency band of 0.12– 0.40 HzEq, calculated in periods of 30 s. The recordings of each session were processed in five stages as described in the above protocol. To compare different sessions, the averages of the observed values of stages 3 and 5 (during and after the hypnoidal state, respectively) were normalized to the average value of segment 1. In stages 2 and 4, corresponding to induction of relaxation in the beginning and at the end of the hypnoidal state, the SC consistently decayed exponentially and was not further characterized.

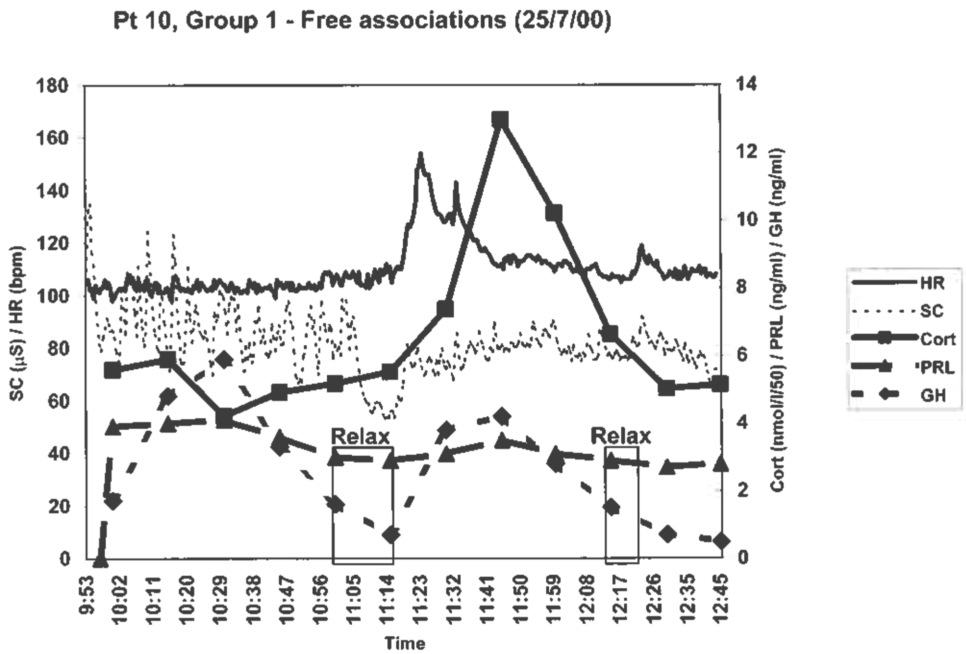

Fig. 1. Hormonal and neurovegetative variables during the first session of a patient who had an emotion of "shock" (see text for definition). Notice the fall in the SC during the first period of relaxation (Relax) and the marked rise in HR, synchronous with the rise in SC, at the end of relaxation period. The serum cortisol concentrations are expressed as nmol/l divided by 50. Notice the intense cortisol surge and the presence of two GH surges, one before and one, synchronous with cortisol, during the hypnoidal state. There are no prolactin surges.

2.3.2. Hormone assays

Blood was drawn every 15 min through an indwelling Butterfly needle placed in a forearm vein and connected to an heparinized tube for the whole duration of the study. Serum cortisol prolactin and VT were measured by conventional methodology: : *PROL-CTK irma SORIN BIOMEDICA*—sensitivity—0.45 ng/ml. Within-assay coefficient of variation for mean values of 22.87 ng/ml was 2.84%. Between-assay coefficients of variation for mean values of 39.55 ng/ml, 92.77 ng/ml and 153.1 ng/ml were 4.0%, 3.6% and 3.9%, respectively; *COAT-A-COUNT cortisol* (RIA, DPC)—sensitivity 6.6 nmol/l; within-assay coefficient of variation for a mean value of 276 nmol/l was 2%; between-assay coefficients of variation for mean values of 31 nmol/l, 129 nmol/l and 519 nmol/l were 7.2%, 4.8% and 9.8%, respectively. *GH IRMA, CIS—Biointernational*—sensitivity 0.1 ng/ml (1st IS 80/505); within-assay coefficients of variation for values of 3.4 ng/ml and 17 ng/ml were 2.4 and 2.8%, respectively. Between assay coefficients of variation for values of 3.3 ng/ml and 16.4 ng/ml were 4.2% and 4.4%.

2.3.3. Analysis of the hormonal data

Cortisol and prolactin peaks were defined according to the general criteria used for the construction of algorithms for the study of episodic secretion (Merriam and Wachter, 1982). As the time series were short (12–15 points) the peaks were identified visually and defined according to the following criteria: For one, two and three or more points the required elevation above baseline was 50%, 35% or 20%, respectively. Incomplete peaks at the end of the session were considered valid for all hormones. A peak of GH was considered to exist when a value equal or superior to 1 ng/ml was observed. The different treatment given to GH was due to the fact that the concentration of this hormone had, usually, stretches of baseline values below the limit of detection of the assay used, before and/or after peaks. Initial values of cortisol or prolactin, on the other hand, were always part of a descending trend corresponding to their respective circadian rhythms.

The baseline for GH was the limit of detection of the assay (0.1 ng/ml). The baseline for the other two hormones was defined by the trend line of the hormonal concentrations after exclusion of the peaks (Merriam and Wachter, 1982). When the trend line was too steep, such that it would project to negative values during the session, the baseline was defined as the value preceding the onset of the surge.

2.3.4. Psychological data

In order to relate the hormonal responses observed during the sessions of free associations to the preceding psychological variables, these were classified, according to three criteria:

- A—*Intensity of the emotion:* 1. Visible emotion (crying, shouting, laughing, wringing hands or restlessness); 2. No visible emotion or emotion expressed only by changes in mimics or in the SC.
- B—*Modes of expression of the associations* during the sessions: 1. Evocation of real events. In this group, the persons involved in the evocations were clearly

identified and the place and time of the events were plausible. All evoked events were found, on debriefing, to have actually occurred; 2. Evocations with defensive elements. In this group, the evocations, even when based in real events, contained elements that could not be identified ("....there is someone in the room but I cannot figure out who he is...") or situations that were not correspondent to reality in terms of place or time of occurrence (e.g., events that occurred at adolescence described as if they had occurred early in childhood). Sometimes, a whole story was "evoked" as if it happened at an implausible time or place (Examples: Middle Ages, Ancient Egypt, the time of "Star Wars"). 3. Defensive mode. Refusal or inability to verbalize meaningful associations. Examples—"I would rather not talk about this". "I do not feel anything", "I only see a yellow light", "Something is upsetting me but I don't know what".

- C—*Type of feeling*: 1. Shock. The patient is struck by the unexpected evocation of a traumatic episode of her past and re-enacts the episode as if it was actually occurring; 2. Intimidation. The patient has a marked rise in the SC concomitant with the arrival of the therapist, persisting as a plateau until the induction of the relaxation. During the session, or on debriefing, it becomes clear that the patient was upset with the upcoming session and with the presence of the therapist; 3. Fear; 4. Guilt or shame; 5. Rage against others or self.; 6. Sadness, abandon or humiliation. No rage expressed; 7. Other or not characterized.

The above classifications were applied by two of three of the members of the research team (LS—endocrinologist with training in psychoanalysis, MS—psychiatrist and LB—psychologist), after each session, before the results of the hormonal tests were available. In eight instances two hormonal surges, separated by 30–45 min, occurred during hypnosis in the same session. For the purpose of the calculations these sessions were split in two segments and the criteria to classify the type and mode of expression of the emotions were applied, a posteriori, to each segment separately.

2.3.5. *Statistics*

One-way ANOVA with the Student-Newman–Keuls correction was used for the comparisons of the means of HR and VT during stages 1, 3 and 5 of each type of sessions. For the SC, due to the wide and biologically irrelevant dispersion of the mean initial values, a different strategy was used. The means of stages 3 and 5 were normalized for the respective stage 1. The z-test was then used for the comparisons with stage 1. Student's t-test was used for the comparisons of the means of stages 3 and 5 (paired test) and of the areas under the curve (AUC) (unpaired test). The two-tailed Fisher's Exact Test was used for the comparison of number of hormonal peaks during the different types of sessions, to establish associations between hormonal peaks in the same sessions and between hormonal peaks and modes of expression and types of emotions. All operations were performed using the Statgraphics Plus software, version 4, Manugistics, Inc., 1998.

3. Results

3.1. First study

3.1.1. Subjective experiences

At the end of the blank sessions all subjects reported that they had a pleasant experience.

The overall experience during the nursing sessions. was described from "pleasant" to "enthralling" by ten of the women. They felt as if they were actually nursing a child and nine of them felt engorged breasts. There were two exceptions: 1. A woman who was tense at the beginning of the session and could never fully relax; 2. A woman who cried and woke up in the middle of the session. On debriefing she told that the attempt to induce a fantasy of pregnancy had been very painful to her because she had faked an unwanted pregnancy in the recent past in order to bring her boyfriend closer to her. As a consequence he had broken up the relationship. Because of this, for the purposes of calculations, this session was considered to be of the "free associations" type and not of the "breast feeding" type.

The sessions of free associations were rich in associations and emotions in all but one case. The exception was a psychologist with experience in this type of interventions. During the session she told a story in dreaming mode and showed no evidence of emotion during the whole session. When the sessions caused painful experiences, as occurred in most subjects, the therapist negotiated with the patient, before the end of the session, a redefinition of her problems and a way (and will) to tackle them. As a result, all enjoyed the experience and stated that they felt more confident and had gained some personal benefit from the session.

One of the volunteers revealed disturbing and unexpected difficulties during the session of free association. She was offered psychotherapeutic follow up.

3.1.2. SC and HR

During the periods of relaxation, at the induction and at the end of the hypnoidal state (stages 2 and 4) there was a marked decline in the SC observable in most sessions, as illustrated in Fig. 1. Conversely, rises were observed at the end of stage 4 in all types of sessions (awakening) and at the end of stage 2 in the sessions of free associations.

During phase 3 of blank and nursing sessions the mean value of the SC was markedly reduced as compared to phases 1 and 5. The actual values are represented in Fig. 2. The HR increased in synchrony with SC during episodes of crying, shouting or laughing in phase 3 of the sessions of free associations. The HR was reduced during phase 3 of the blank and "breast feeding" sessions as compared to phases 1 or 5 but the differences did not reach significance. The VT did not show any change as a function of the phase or type of the session.

3.1.3. Hormone concentrations in blood

There were two cortisol, two prolactin and two GH surges in the 12 blank sessions and one cortisol, one prolactin and two GH surges in the 12 "breast feeding" sessions.

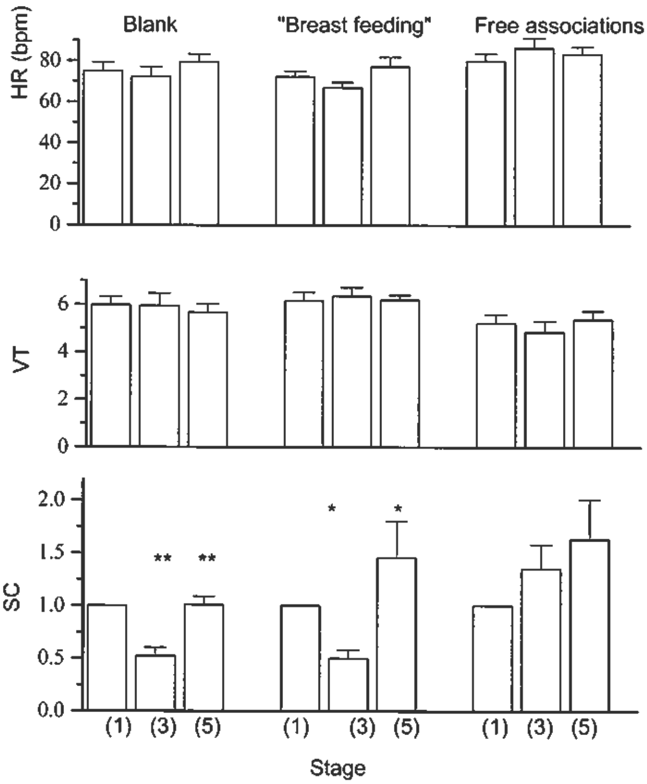

Fig. 2. Mean values of HR, VT and SC during stages 1, 3 and 5 according to the type of session. The SC is expressed as a fraction of averaged stage 1. The error bars represent the standard error of the mean. The "p" values for stage 3 represent the significance of the difference for stage 1; the "p" values for stage 5 represent the difference for stage 3. ($*$ - $p < 0.01$; $**$ - $p > 0.001$).

By contrast, during the 17 sessions of free associations there were eight cortisol, seven prolactin and nine GH surges (Fig. 3). The number of cortisol, prolactin and GH surges during the free associations sessions was significantly higher than those in the blank plus "breast feeding" sessions (p values < 0.03, <0.05 and <0.02, respectively).

3.2. Second study

3.2.1. Hormone concentrations in blood

The observations in the 38 sessions of free associations in this study were pooled with the 17 sessions of free associations from the first study in an attempt to match psychological observations and hormonal responses.

Number of hormonal surges preceding and during the hypnoidal state (stage 3)—A total of 32 cortisol, 18 prolactin and 28 GH surges was observed during stage 3 in the 55 sessions studied. The number of hormonal surges occurring during the 60

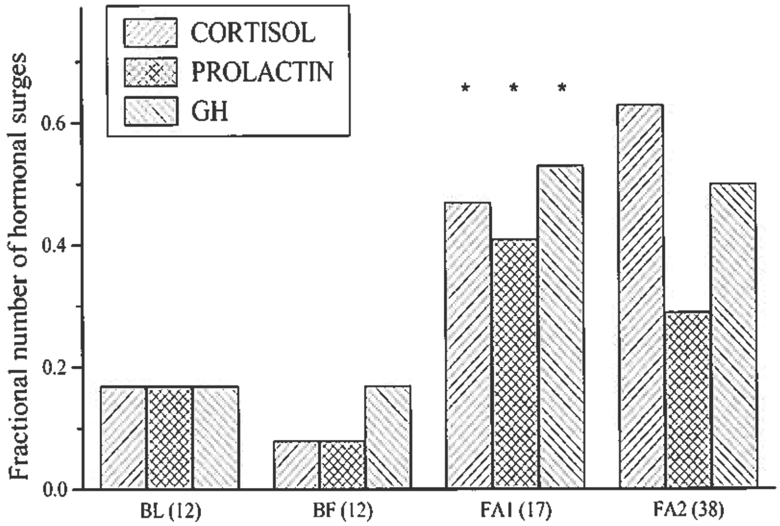

Fig. 3. Fractional number of hormonal surges during stage 3 according to the type of sessions. BL – blank sessions; BF – "breast feeding" sessions; FA1 – sessions of free associations (first study); FA2 – sessions of free associations (second study). In brackets ( ) – number of sessions studied. The "p" value ($* <$ < 0.05) represents the significance of the differences between the blank and "breast feeding" sessions and the sessions of free associations of the first study (FA1). FA2 is represented to illustrate its similarity to FA1.

minutes preceding the induction (i.e., during stage 1) was compared to those during stage 3 as a further control for spontaneous activity. The results were the following: cortisol—3/55 v. 32/55 ($p < 0.001$); prolactin −3/55 v. 18/55 ($p < 0.001$); GH—18/55 v. 28/55 ($p<0.1$).

During stage 3 there were significantly more cortisol than prolactin surges (32/55 v. 18/55; $p <0.02$). The number of GH surges (28/55) was intermediate and not significantly different from either cortisol or prolactin's.

Associations between hormonal surges—Cortisol and prolactin surges were negatively associated ($p < 0.03$). GH had no significant association either with cortisol or with prolactin.

Hormonal peaks and intensity of emotions—There were visible emotions in 37 of the sessions. Visible emotions were positively associated with GH surges ($p < 0.05$). There were no significant associations with cortisol or prolactin

Hormonal peaks and mode of expression of emotions—The number of cortisol surges was associated with evocations of real events ($p < 0.01$). Conversely, the number of cortisol surges was negatively associated with the group of "evocations with defensive elements" alone ($p<0.01$), or pooled with the group of "defensive mode" ($p<0.01$). The number of sessions in defensive mode was too small (three cases) for a meaningful analysis of this group alone. No significant correlations were observed between prolactin or GH surges and modes of expression of emotions.

Hormonal peaks and type of emotions—The number of cortisol surges was significantly associated with the pooled group of "shock" and "intimidation" ($p < 0.02$).

Cortisol peaked in every instance of shock (two sessions) and intimidation (five sessions). The intensity of these surges was of the highest observed, as shown in Fig. 4. The values of the AUC of the categories "shock" and "intimidation" pooled together were significantly higher than those of the pool of all the other categories (12.4 μmol.min.l^{-1} v. 7.1 μmol.min.l^{-1}; $p < 0.005$). Rage was negatively associated with cortisol ($p = 0.05$) and had a marginal positive association with prolactin ($p <$ 0.07). The distribution of GH surges did not show any significant association with types of emotions.

4. Discussion and conclusions

This work reports on hormonal and neurovegetative events temporally associated with emotions induced under hypnosis. This modified state of consciousness allows the induction of actively imagined fantasies, such as the fantasy of breast feeding, as well as the evocation of memories, both utilized in the present study. During this state the subjects maintained contact with external reality and retained full memory of their subjective experiences. This experimental approach was selected for the following two reasons: 1. Because it allowed to test whether a subjective state of parenthood and breast feeding might elicit a prolactin response, as suggested by previous studies in humans and primates (Auerbach and Avery, 1981; Dixon and George, 1982; Sobrinho and Almeida-Costa, 1992; Ziegler et al., 1996); 2. Because the emotions elicited in sessions of free associations about themes selected by the patients themselves were easy to obtain, diverse, often intense, and could be charac-

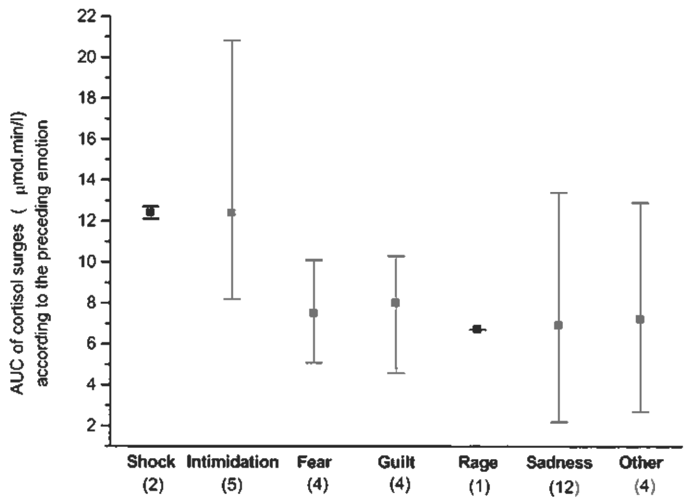

Fig. 4. Intensity of the cortisol surges, expressed as AUC (mol.min.l^{-1}) as a function of the the type of the preceding emotion. The solid squares ■ represent the average values. The error bars represent the minimal and maximal values of range.

terized in a biographical context. Therefore, they allowed the possibility of matching characteristics of emotions with hormonal responses.

During the sessions, SC, HR and VT were monitored. The SC, is a sensitive and non-specific marker of arousal. During the relaxation period (stage 2), the SC fell exponentially in most sessions indicating that the sympathetic activity was being markedly reduced as compared to baseline levels. During the blank and "breast feeding" sessions the SC was reduced to about half of its initial value during the whole session. This reduction was not observed during the sessions of free associations, in accordance with the state of arousal thus obtained.

The HR changes were parallel to those of SC but much less intense. In the presence of visible emotions there were, usually, spikes of HR synchronous with that of SC (Fig. 1).

The VT, as evaluated by the spectral analysis of the HR variability (Porges, 1995), did not vary either with the stage or type of session. The absence of variation with stage was expected during the sessions of free associations as the subjects were talking, and the respiration erratic. However, during the blank and breast-feeding sessions the values of VT during stage 3 were also similar to those of stages 1 or 5, despite the fact that the patients were not talking and had a deep, regular, respiratory rhythm most of the time. This unexpected observation suggested that the relaxation and reduced sympathetic activity obtained was not associated with any meaningful change in the parasympathetic tone, as evaluated by the method used.

As hormonal surges occur spontaneously in normal, free-moving, volunteers at the average rate of 19/24 hours for cortisol (Veldhuis et al., 1989), 20/24 hours for prolactin (Veldhuis et al., 1994) and 11/24 hours for GH (Veldhuis et al., 1994), the first study was designed such that the initial session consisted of the induction of a pleasant, relaxing, state to be used as a blank control. In these sessions few hormonal surges were observed during stage 3.

The "virtual breast feeding" sessions were successful, in most subjects, in inducing a subjective state of motherhood and a fantasy of nursing felt as real, including a feeling of engorged breasts and bodily modifications. Yet, the hormonal pattern in these sessions was similar to that of the blank sessions. With the caveat that neither the setting of the experiment nor the duration of the "delusional" state of breast feeding reproduce the spontaneous condition of pseudopregnancy, our observations do not support the hypothesis that psychogenic factors stimulate prolactin secretion through the induction of a psychological state of motherhood and nursing, as has been suggested (Sobrinho and Almeida-Costa, 1992).

The sessions of free associations differed from the blank and nursing sessions in that there were significantly more hormonal surges than in blank and "breast feeding" sessions. This was true for cortisol, prolactin and GH. Therefore, all these hormones respond to psychological stress in humans

The analysis of the hormonal surges during all the 55 sessions of free associations demonstrated that cortisol surges were significantly more common than prolactin's (32/55 v. 18/55; $p < 0.02$). An important finding was the negative association between the occurrence of prolactin and of cortisol surges indicating that the prolactin response is an alternative to, rather than an extension of, the more common cortisol response. This negative association is in accordance with the data by Gerra et al. (2001) who observed that volunteers submitted to psychological stress (Stroop color word interference task plus public speaking plus a mental arithmetic test in front of an audience) responded with an increase in ACTH and cortisol *and* a decrease in prolactin serum levels.

In rodents, lesion studies demonstrate that the neural pathways responsible for cortisol and prolactin responses to stress are different (Van de Van de Kar and Blair (1999). However, we are not aware of any observations suggesting that the two hormonal responses can be dissociated according to the stimulus. Instead, prolactin surges are often considered as a part, if often absent in humans, of the overall stress response. Acute prolactin responses to "psychological" stress are commonplace in rodents (Ratner et al., 1989) but have been documented only sporadically in humans (Biondi and Picardi, 1999). An interesting observation was published by Reichlin (1988) who described a marked prolactin surge (from 40 ng/ml to 180 ng/ml) in a pregnant patient while arguing with the attending nurse during the course of a metabolic study.

Before postulating qualitative differences between species it must be considered that the aggressions incurred by humans who volunteer for experiences of psychological stress are not comparable, in nature or intensity, to the stressors applied to rodents in experimental protocols—restraint, foot-shock, etc..

Surges of GH during the sessions of free associations were almost as common as those of cortisol (28/55 v. 32/55), in agreement with reports describing GH as an hormone of psychological stress (Kosten et al., 1984; Gerra et al., 1996). However, GH surges were also frequent during stage 1 of the sessions, i.e., before the induction of the free associations, in striking contrast with the almost absence of cortisol or prolactin surges during this period. It is possible that the high number of GH surges during stage 1 reflects the sensitivity of this hormone to environmental stimuli (manipulation for the placement of electrodes, puncture of the vein, etc.) to which cortisol and prolactin appear to be insensitive. GH also differed from cortisol and prolactin in that its surges were positively associated with the intensity of the emotions. It is, thus, possible that GH may be primarily related to the motor component of the emotions while cortisol and prolactin may be more related to the associated feelings or mechanisms of retrieval of memories. The fact that the release of cortisol is associated positively with the evocation of real events and negatively with evocations containing defensive elements suggests that cortisol surges are likely when evocation of the emotions are linked with the evocation of the correspondent events. Conversely, when evoked emotions, however intense, cannot be clearly associated with the accompanying events, the occurrence of cortisol surges is unlikely.

The attempt to match the type of the emotions to hormonal responses is difficult for, at least, two reasons: 1. the identification of types of emotions, even when interpreted in a biographical context, contains a degree of subjectivity; 2. the time course of the emotions, that can be quite fleeting in one single session, does not necessarily coincide with that of hormone concentrations measured at fixed intervals and conditioned by their distribution volumes and clearance rates.

Due to the above constraints an unequivocal correspondence between hormonal surges and types of emotions could only be made in the minority of cases in which the latter were obvious and prolonged as were the cases of shock and a few others. More often, however, the sessions consisted of evocations of different episodes. As a consequence, the "type" of the emotions reflected the overall assessment of the dominant emotion expressed throughout the session and no claim can be made as to the "purity" of the emotions observed. As a consequence of this within-session variability in the emotional states, the differences in the mean HR during stages 1, 2 and 3 of the different types of sessions did not reach significance, even though marked (but short-lived) spikes were observed in association with intense emotions. Neurophysiological correlates and cerebral mapping of single, "pure", emotions have been studied in a different setting (Damasio et al., 2000). However, because of the very design of these studies, the time-course of the observations was very short (less than one minute) and no hormonal variables were studied. Despite these difficulties our observations suggest that the hormonal responses to psychological stress may be related to the type of emotion elicited. Cortisol was associated positively with "shock" and "intimidation" ($p<0.03$) and negatively with rage ($p< 0.05$). Therefore, our observations are in accordance with current thinking that cortisol, is *the* hormone of the psychological stress of threatening situations. Fear would also be expected to be associated with cortisol. However, as "shock" and "intimidation" were considered as separate entities because they could be objectively defined, "fear" became a residual category. Prolactin, on the other hand, had a marginal positive association with rage ($p<0.07$). No other associations between hormonal surges and types of emotions were observed. The above described association of some emotions with specific hormonal responses together with the negative correlation between cortisol and prolactin surges are in accordance with the formulation that different emotions are associated with different body states expressed as different patterns of endocrine, and also neural (Damasio et al., 2000), activity (Damasio, 1994).

The role of characteristics of the subjects in the hormonal responses to psychological stresses was not addressed in the present study. However, there is evidence, both from animal and human studies, that early losses (Sobrinho et al., 1984; Luecken, 1998) or early manipulations (Liu et al., 1997), dismissing attachment (Scheidt et al., 2000), coping styles and personality traits (Kirschbaum et al., 1995; Biondi and Picardi, 1999) and manual preference (Martins et al., 2001) or "pawness" (Waters et al., 1996) may condition the hormonal responses to psychological stress.

In conclusion, the present study provides evidence that cortisol, prolactin and GH respond to psychological stress in humans. However, they appear to be regulated differently from one another and do not behave as components of a generalized "stress" response. Cortisol responds consistently and intensely to shock and intimidation. Prolactin surges are observed less frequently and are negatively correlated with cortisol, behaving as an alternative response. Prolactin surges are not stimulated by a fantasy of breast feeding but may be associated with the feeling of rage. Growth hormone surges are common, both before and during the sessions and are associated with the intensity, but not with the type, of the emotions, suggesting that this hormone may depend, in a non-specific way, on the motor component of the emotions.

It is possible that a larger number of observations may reveal patterns of association between emotions and hormones that have been, so far, inconspicuous. It is also possible that the way the emotions were classified by intensity, mode of evocation and type is not the best to capture the characteristics that determine the hormonal surges. Despite these limitations, it is our belief that the present study provides useful information for the analysis of the diversity of hormonal changes observed in psychological stress.

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