SERUM MELATONIN CONCENTRATION IN THE CHILD WITH NON-ORGANIC FAILURE TO THRIVE: COMPARISON WITH OTHERS TYPES OF STRESS

A. MUÑOZ-HOYOS, A. MOLINA-CARBALLO, J. UBEROS, F. CONTRERAS-CHOVA, M. DEL CARMEN AUGUSTIN-MORALES, M. RUIZ-ALBA, and G. GALDÓ-MUÑOZ

Unidad de Gestión Clínica de Pediatría, Hospital Universitario San Cecilio de Granada, Universidad de Granada, Granada, Spain

Received August 20, 2008 – Accepted January 28, 2009

Human beings must adapt both to novel, unfavourable conditions and to circumstances of physical or psychological isolation. The initial response to stress depends fundamentally on the activation of the HPA axis. In regaining homeostatic equilibrium, melatonin plays a role due to its synchronising and anti-stress properties. To study the role of melatonin and the pineal gland in the organic and/or behavioural response to acute or chronic stress, 311 children were divided into two large groups: 1) Control Group - 121 healthy children classified, in turn, into 4 control subgroups, one for each pathology being studied; 2) Problem Groups, classified as *traumatic stress* (n=58), *surgical stress* (n=38), *psychic stress* (n=64) and *febrile stress* (n=30), according to pre-established clinical criteria. These groups were sub-classified according to the degree (low or high) and duration (acute or chronic) of the stress. This study used a case controlled, cross sectional design. Serum melatonin was measured by radioimmunoassay (RIA). In all the situations of acute stress, melatonin increased at a rate directly proportional to the severity and/or duration of the stress-causing stimulus. In contrast, in chronic stress, i.e. the Affective Deprivation Syndrome (or Psychological Dwarfism) with or without non-organic failure to thrive, resulted in the opposite response with a significant reduction of melatonin. In conclusion, in acute stress an increase in the bioavailability of melatonin could contribute to maintaining homeostatic balance. The lack of an appropriate response to acute stress could make some groups of patients (Affective deprivation syndrome with or without growth failure) predisposed to suffer depressive symptoms associated with a wide range of neurological, endocrinological or immunological consequences.

Human beings must adapt to novel, unfavourable conditions or to circumstances of physical or psychological isolation. This homeostatic response is known as the stress reaction (1). Glucocorticoids and cytokines are important mediators of injury responses (2). It has been suggested that glucocorticoids and central/peripheral cytokines are finely balanced to maintain homeostasis with reciprocal inhibition/activation pathways being present (3). Because of their potent activatory effect on the HPA axis, proinflammatory cytokines could contribute to the elevated levels of glucocorticoids that are observed

Key words: melatonin, children, acute stress, febrile stress, surgery stress, traumatic stress, affective deprivation syndrome, non-organic failure to thrive syndrome, low melatonin syndrome

Mailing address: Dr. D. Antonio Molina-Carballo,
Departamento de Pediatría,
Facultad de Medicina,
Avda. de Madrid 11, E
18012 Granada, Spain
Tel: ++34 958023996. Fax: ++34 958240740.
e-mail: amolinac@ugr.es

0393-974X (2009)

after stress. Reciprocally, activation of glucocorticoid receptors leads to down-regulation of peripheral and central expression of proinflammatory cytokines, via modulation of other transcription factors, such as AP-1 (a heterodimer of the oncoproteins c-Fos and c-Jun) and NFkB (3). This complex balance appears critical and, if not maintained, it can have clinical consequences (4) in the form of neuronal death or reduced brain plasticity (2).

In humans, the intake of tryptophan causes an important increase in the concentration of melatonin but does not affect that of cortisol (5). In healthy children, the nocturnal administration of tryptophan or of pyridoxine (6) also increases melatonin secretion.

Melatonin is secreted nocturnally by the pineal gland in all animal species, and it has anti-stress properties that are partly attributed to its modulatory function within the central nervous system (7). It presents both hormonal properties regulated by a receptor, like any standard hormone, and also non-receptor mediated properties, that make it capable of being readily disseminated to any organic compartment. Although melatonin has lately been identified in a large number of extrapineal sites, its potential biological actions have not yet been studied. Human lymphoid cells are an important physiological source of melatonin and this melatonin could be involved in the regulation of the human immune system (8-9), possibly by acting as an intracrine, autocrine, and/or paracrine substance. Overall, melatonin acts as a synchroniser of biological rhythms, as an immunomodulator and as an antioxidant (10).

With this in mind, the present study intends to identify the modifications in the circulating concentration of melatonin in children exposed to different types of stress (febrile, surgical, traumatic and psychological).

MATERIALS AND METHODS

This study was comprised of 311 consecutive paediatric patients attending our university-based Hospital. Informed consent was obtained from all parents or guardians in accordance with the care standards established by the Hospital's Institutional Review Board (IRB). The hospital's regular light-dark schedule (lights on from 8 a.m. to 9 p.m.) was observed, with an ambient illuminance between 300–450 lux when the single serum samples for each patient was taken.

In order to meet the objectives established, the subjects were classified according to two criteria: the nature of the stress-provoking stimulus and its intensity. Because the samples of the different experimental groups were collected in separate scheduled periods, samples from the control groups were obtained at the same time as the match in the experimental group. In the experimental design, the groups and subgroups (Table I) were structured as follows:

Febrile stress – N= 30 (9.64% of the total sample). As a Control Group of the patients with this type of stress, we included paediatric patients with no previous perinatal, family or personal history of neurological or endocrine-metabolic disease. To meet criteria as a control subject, the child either had no pathology or only a mild pathology. This group of 22 children were of a similar age and sex to that of the other study groups. In this study samples were taken regularly throughout the day (except during the period from 1 a.m. to 6 a.m.). The blood sample was taken from the patients in the control group at the same time (± 1 hour) as the sample from the age- and gender- matched controls in one of the problem groups.

Surgical stress – N= 76. This group was divided into three subgroups: a) Control group, with 38 normal children, with an average age of 6.64±2.33 years, treated for a mild pathology; b) low-level surgical-stress group (n=23), with an average age of 6.94±3.65 years, similar to that of the control group; c) subjected to high levels of surgical stress group (n=15), with an average age of 7.33±3.35 years, who needed major or complex surgery. This latter group was defined according to a widely accepted classification (11), corresponding to a degree of surgical stress exceeding 5 points; whereas, the low level surgical stress group had a score of $\leq$5 points.

Traumatic stress – N= 88. This group included three subgroups: a) control group, with 30 normal children, treated by us for a mild complaint, and with an average age very similar to that of the other two groups (5.3±3.9 years); b) mild traumatism group, comprising 30 children who attended after suffering

a mild traumatism (requiring neither anaesthesia nor surgery); c) severe traumatism group, 28 children, aged 6.1±2.8 years (without statistical differences compared to the others groups), who required admittance to hospital and continuing observation after suffering a severe traumatic episode.

Psychic stress - made up of 95 patients, aged 4-14 years, and divided in turn into three groups: a) control group, with 31 paediatric patients, of similar age and gender to those of institutionalised patients and those presenting non-organic failure to thrive syndrome, with the following characteristics in common: absence of hereditary or neurological diseases, birth by normal delivery, no significant history of medical illness, normal neuromotor development, and age appropriate educational progress; b) institutionalised group (n= 64) , aged 4-14 years derived from the whole population who were admitted to a social-charitable institution during a prolonged interval (mean time of 34.73 months, with a range from 3 months to 9 years), due to the existence of severe family conflict, abandonment, orphanhood, or similar circumstances. In order to meet the proposed objectives, and because the clinical situation was well-defined (weight and height below the third percentile, no apparent organic cause for growth discrepancies, and the presence of psycho-affective disorders) we further subdivided this group of institutionalised patients (n=41) from the those affected by non-Organic Failure to Thrive syndrome (n=23), psychological characteristics were evaluated by CDS [Children Depression State (Tisher y Lang,1974)] and STAI [State-Trait Anxiety Inventory (Spielberger, Gorsuch, & Lushene, 1970)] tests for all patients in the psychic stress group. Samples for this study group were taken at 9 a.m.

Clinical method

The differences between the various clinical situations examined in this study required the inclusion of the particular characteristics of each study group. All the patients had a common study protocol that included a complete clinical examination and a full case history. In all cases, both the children involved and their parents or guardians were informed verbally of the nature of the study, and written consent was obtained prior to any procedures carried out.

Treatment was individualised according to the differential characteristics of each of the pathologies presented. However, for only some of the patients included in the 'psychic stress' group was it possible to obtain family and personal details from the parent or guardian. In the other cases, the data were obtained from the tutors. This included aspects of interest that had occurred during the years of residence within the institution for the whole group. These patients were given two psychometric tests to analyse their degree of intellectual attainment: the children aged under 8 did the Boehm test of basic concepts and were evaluated on the Columbia Mental Maturity Scale; those aged 8 years and older completed Raven's test of progressive matrices and the Elementary Intelligence Test. An anthropometric and nutritional study was also carried out, to determine dietary aspects that might have bearing on the presence of absence of failure to thrive. This study included the following parameters: weight, height, body mass index, weight/height ratio$_3$, weight index, skinfolds, body density, percentage of body fat, and weight of body fat (in grams).

After the surgical intervention, the patients with surgical stress were classified into two subgroups, in accordance with the Anand and Aynsley-Green scale (11) (Table II), as either suffering a low level of surgical stress (≤ 5 points on the latter scale: traumatisms not requiring anesthesia or surgery) or a high level of surgical stress (>5 points). This assessment included the amount of blood volume losses, the site of the intervention, the characteristics of the dissection, the visceral trauma from the process, the duration of the intervention, and other associated factors (11).

The patients with traumatic stress were divided into two subgroups (mild traumatism, severe traumatism) exclusively on the basis of the type of traumatism suffered and on the definitive assessment of the traumatologist.

The measurement of melatonin was carried out by a radioimmunoassay technique, using two different commercially available RIAs (Euro-Diagnostics, Apeldoorn, The Netherlands, for the samples of the Psychic stress group and DLD, Hamburg, Germany, for the other groups). Quality control was performed. The cross-reactivity of the Euro-Diagnostics kit was less than 1% for related substances; with an interassay variability of 9.5% at a

concentration of 35 pg/ml. The DLD kit shows intra- and interassay coefficients of variation of 11.3% and 6.3%, respectively. The recovery of added melatonin was 84.4%, and the sensitivity of the assay was 0.02 nmol/L; this kit presents an antibody that is very specific for melatonin, and virtually no cross-reaction with its principal metabolites (6-hydroxy-melatonin and 6-sulphatoxy-melatonin) was present.

Statistical methodology

The following tests were carried out: comparison of the means, one way analysis of the variance with subsequent pair-wise comparison (using the Bonferroni test, as the study groups had different numbers of subjects) when statistical significance was detected, and, finally, a simple linear correlation analysis.

RESULTS

Febrile stress

The serum melatonin values for the control group (n= 22) were 26.54±9.57 pg/ml. Of the 52 patients in this group, the mean value was notably higher: 49.33±12.70 pg/ml. A simple rise in body temperature was considered a cause of neurological stress that was tolerated well by the patient, provided it was not associated with any type of clinical complication. Consequently, this study group was established as a complement to previous studies of the pineal response to febrile or epileptic convulsions.

Surgical stress

The comparative analysis of melatonin levels in each of the study groups provided the following results: control group (CG) 29.84±10.68 pg/ml; low surgical stress (LSS) 43.39±12.56 pg/ml; high surgical stress (HSS) 56.66±19.71 pg/ml. The F_{exp} values were 23.06 ($p<0.001$) and the subsequent ANOVA values between the groups were significant: CG/LSS (T_{exp} = 4.05; $p<0.01$; CG/HSS (T_{exp} = 6.45; $p<0.001$). Finally, between the two types of stress HSS/LSS, T_{exp} = 2.94; $p<0.05$.

In the control group, there was found to be a certain degree of linear negative correlation between the variables 'age' and 'aMT concentration' (r=-0.33). This relation was considerably more marked in relation to the low surgical stress group (r=-0.79; $p<0.001$) and less so as the degree of stress increased (r=-0.25; p=NS).

With respect to the analysis of the correlation between the variables 'degree of stress' and 'aMT concentration', there was shown to be a positive linear relation, as follows: HSS/aMT: r=0.44; $p< 0.05$; LSS/aMT: r= 0.73; $p<0.01$. Total patients subjected to surgical stress / aMT: r=0.55; $p<0.01$. The correlation between the degree of surgical stress and the serum concentration of melatonin is reflected in the 'high surgical stress' group with a Pearson 'r' score of 0.44 ($p<0.05$) and a regression slope of y=27.7+2.72x. The respective values corresponding to the low stress group were as follows: r=0.73, $p<0.01$, and regression slope y=9.4+11.1x. Finally, for the whole group subjected to surgical stress, the respective values were r= 0.55, $p< 0.01$ and regression slope y=35.5+2.17x.

Traumatic stress

Of a total of 88 paediatric patients, 30 cases were controls, and the rest were divided into two categories, mild traumatisms (n=30) and severe traumatisms (n=28). The aMT values obtained were 30.13±11.41 pg/ml in the control group, 33.65±10.40 pg/ml in the mild traumatism group and 82.43±17.12 pg/ml in the severe traumatism group. The comparative study produced highly significant results (F_{exp} 140.2, $p<0.001$), with the highest values being found in the contrast between the control group and that of severe traumatisms (T_{exp}=15.1, $p<0.0001$).

Psychological stress

This situation of stress produced the greatest differences with the control group, but in the inverse sense (F_{exp}=35.87; $p<0.001$), as the highest plasma values corresponded to the control group, 39.48±5.95, and the values were notably lower in the other two groups,

Table I. *Human subjects: groups of patients and number of cases per study group.*

Pathology Subgroups	No. of Cases
Febrile Stress	
Control	22
Febrile Group	56
Surgical stress	
Control	38
Low stress	23
High stress	15
Traumatic stress	
Control	30
Mild traumatism	30
Severe traumatism	28
Psychic Stress	
Control	31
Institutionalised	41
NOFT	23

Table II. *Evaluation scale for degree of Surgical Stress.*

Criteria	Score
Volaemia losses	
< 5%	0
5-10%	1
10-15%	2
>15%	3
Location	
Superficial	0
Intra-abdominal	2
Intra-thoracic	3
Superficial Dissection (skin, muscles, etc.)	
Minimal	1
Moderate	2
Maximum	3
Visceral trauma	
Brief	1
Prolonged	2
Minor resection	3
Major resection	4
Duration of surgery (minutes)	
< 30	1
30-90	2
90-180	3
180-300	4
> 300	5
Associated factors	
Hypothermia	
1.5 - 3.0ºC	1
> 3.0 ºC	2
Profound	3
Infection	
Localised	1
Generalised	2
Prematurity	
37-34 weeks	1
< 34 weeks	2

Adapted from Anand and Aynsley-Green 1988

25.79±7.97 and 25.85±7.76 (pg/ml), respectively. In later comparisons, the analysis of the variance revealed differences between the control group and the other two, with experimental values of T_{exp}=18.37 ($p<0.0001$) for the group of institutionalised patients, and T_{exp}=22.8 ($p<0.0001$) for the institutionalised patients suffering non-organic failure to thrive (Fig. 1).

DISCUSSION

The present paper describes changes in the circulating concentration of melatonin under different situations of stress that are frequently encountered among paediatric patients. In the human neonatal period the high melatonin production (12) is transient, does not appear to be controlled by light, and does not present an endogenous circadian rhythm (13). An endogenous circadian rhythm develops many months later and may serve antioxidant functions (13). These high melatonin levels seem to derive from extrapineal organs, suggesting that during the response to an injury the production of melatonin can be transiently shifted from an endocrine (pineal) to a paracrine (immunocompetent cells) source (14).

Thus, when a paediatric patient has a fever, we show that the circulating concentration of melatonin is higher than normal (49.33±12.79 pg/ml *vs* 26.54±9.57 pg/ml in the control group) (Fig. 1), a situation of organic stress which is associated with greater melatonin secretion. The data from all the study groups and the statistical comparisons between the different subgroups are shown in Fig. 1. This figure also shows that the melatonin response is proportional to the intensity and duration of the stress. These observations have been reported in other studies (15-16). As limitations of this study, we recognize that samples of the study groups, except the febrile seizure group, were taken in the morning and therefore may not adequately reflect the nocturnal pineal secretion.

In contrast with the above observations, in a situation of mild to moderate chronic stress, adaptive neuroendocrine changes take place in the opposite direction (17). This is shown by the data we present for a model of chronic stress of psychosocial origin, namely the Affective Deprivation Syndrome (or Psychological Dwarfism), with or without non-organic failure to thrive (18). Abandonment and/or different types of deprivation lead to many patients being admitted to an institution, and effectively separated from their parents. Thus, in non-organic failure to thrive (19), which may be considered the

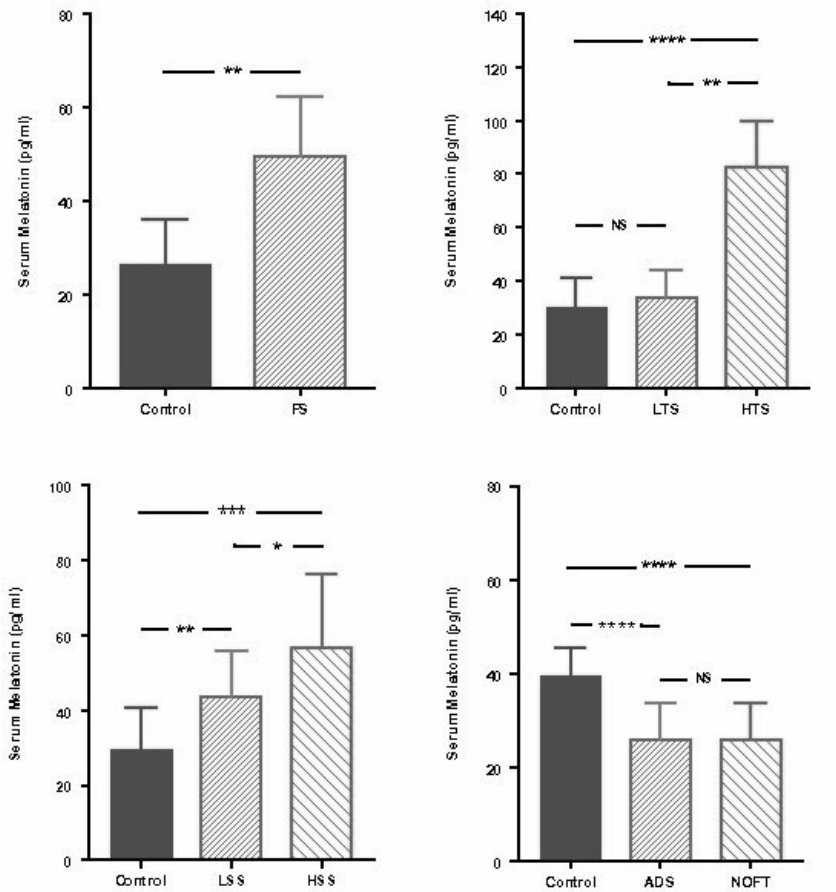

Fig. 1. *Morning serum concentrations of melatonin under the different types of stress studied: Febrile stress (FS), Surgical stress (low: LSS; high: HSS), Traumatic stress (low: LTS; high: HTS) and Psychic stress (Affective deprivation syndrome: ADS, Non-organic Failure to thrive syndrome: NOFT). Each type of stress is compared with the specific control group for healthy children of similar age, sex and weight, evaluated simultaneously and in surroundings of comparable characteristics. Values are expressed as mean ± standard deviation. NS: Differences not statistically significant, *= p <0.05, **= p<0.01; ***= p<0.001; ****= p<0.0001.*

most severe expression of the Affective Deprivation Syndrome, the serum concentration of melatonin is at a minimum, although the difference between the two psychological stress groups is not statistically significant.

Furthermore, among a group of adults who spent a year in Antarctica (exposed to social isolation and to extreme environmental conditions), a statistically significant decrease in the concentration of pituitary stress hormones and of melatonin was noted compared to control subjects (20). Although these were healthy adults who participated voluntarily in the experiment; some similarities exist between those adult subjects and children in our study who were in the Affective Deprivation Syndrome (with or without failure to thrive). In both cases, there is a deficit of social interaction, and the response is the same in each case. The fall in hormonal levels seen in both scenarios may be the expression of adaptive neuroendocrine response to chronic stress.

In healthy adults, acute acoustic stress is associated with a significant increase in the excretion of cortisol and melatonin, while the administration of the anti-psychotic quetiapine reduces the nocturnal secretion

of cortisol without affecting that of melatonin. This highlights that for people with no psychiatric disorder, the activity of the HPA axis may be fluctuating compared to patients with psychic illness (21). Also in adult patients it has been reported that some types of depression may be related to reduced levels of melatonin (22), and that an initial reaction to stress combined with the development of resistance to corticotropin releasing hormone may be related to a low-melatonin syndrome in some patients suffering with depression (23). In this way, among breast-fed infants, the administration of vaccines provokes an increase in cortisolaemia, in inverse proportion to the patient's age (24). In a significant minority of these patients, however, the opposite occurs and cortisolaemia levels fall. These latter patients could be susceptible to the development of a depressive state if, before they attain a sufficiently mature HPA axis, they are subjected to constant psychological stress. This especially may be the case if the response to CRH could be shown to be regulated by psycho-environmental variables (25).

The physiological effects of the activation of the HPA axis and of the pineal gland are opposed, with the latter secreting melatonin and producing an anti-stress effect (26). Several studies have reported that chronic stress has an impact on the areas of the brain implicated in learning and in emotional responses (27-28). Also, a melatonin deficit may explain, in part, that the children who suffer abuse have greater sleep latency, reduced sleep efficiency and present a higher proportion of motor activity during the night than normal children or those suffering depression (29). Different models of stress have demonstrated the utility of treatment with melatonin (30-33).

In summary, in acute stress an increase in the bioavailability of melatonin could collaborate in maintaining homeostatic balance and help overcome the stress-provoking situation without adding further organic deterioration. The lack of an appropriate response, i.e. the organic incapability of confronting stress on a genetic basis, and/or the fact of repeated stress, from exhaustion of the homeostatic mechanisms, could make some groups of patients liable to suffer depressive symptoms associated with a wide range of prejudicial consequences in the neurological field (delayed learning), in the endocrine system (delayed growth) or in the immune system (repeat infections). All these pathologies are present in Affective Deprivation Syndrome (or Psychological Dwarfism).

ACKNOWLEDGEMENTS

The authors thank Mr. Glenn Harding for translating the original manuscript.

REFERENCES

1. Lightman SL. The neuroendocrinology of stress: a never ending story. J Neuroendocrinol 2008; 20:880-4.
2. Rothwell NJ. Annual review prize lecture cytokines - killers in the brain? J Physiol 1999; 514:3-17.
3. Webster JI, Tonelli L, Sternberg EM. Neuroendocrine regulation of immunity. Annu Rev Immunol 2002; 20:125-63.
4. Bugental DB, Martorell GA, Barraza V. The hormonal costs of subtle forms of infant maltreatment. Horm Behav 2003; 43:237-44.
5. Hajak G, Rodenbeck A, Ehrenthal HD, et al. No evidence for a physiological coupling between melatonin and glucocorticoids. Psychopharmacology (Berl) 1997; 133:313-22.
6 Muñoz-Hoyos A, Amorós-Rodríguez I, Molina-Carballo A, Uberos-Fernández J, Acuña-Castroviejo D. Pineal response after pyridoxine test in children. J Neural Transm Gen Sect 1996; 103:833-42.
7. Maestroni GJM, Conti A. Anti-stress role of the melatonin-immuno-opioid network: Evidence for a physiological mechanism involving T cell-derived, immunoreactive b-endorphin and met-enkephalin binding to thymic opioid receptors. Int J Neurosci 1991; 61:289-98.
8. Carrillo-Vico A, Lardone PJ, Fernandez-Santos JM, et al. Human lymphocyte-synthesized melatonin is involved in the regulation of the interleukin-2/interleukin-2 receptor system. J Clin Endocrinol Metab 2005; 90:992-1000.
9. Muñoz-Hoyos A, Espín-Quirantes C, Molina-Carballo A, et al. Neuroendocrine and circadian aspects (melatonin and ß-endorphins) of atopic dermatitis in the child. Pediat Allergy Immunol 2007; 18:679-86.

10. Acuña-Castroviejo D, Escames G, Macías M, et al. Cell protective role of melatonin in the brain. J Pineal Res 1995; 19:57-63.
11. Anand KJ, Aynsley-Green A. Measuring the severity of surgical stress in newborn infants. J Pediatr Surg 1988; 23:297-305.
12. Muñoz-Hoyos A, Bonillo-Perales A, Avila-Villegas R, et al. Levels of melatonin in the first week of life: its relationship with the antioxidant response in the perinatal period. Neonatology 2007; 92:209-16.
13. Tan DX, Manchester LC, Sainz RM, Mayo JC, Leon J, Reiter RJ. Physiological ischemia/reperfusion phenomena and their relation to endogenous melatonin production: a hypothesis. Endocrine 2005; 27:149-58.
14. Pontes GN, Cardoso EC, Carneiro-Sampaio MM, Markus RP. Injury switches melatonin production source from endocrine (pineal) to paracrine (phagocytes) - melatonin in human colostrum and colostrum phagocytes. J Pineal Res 2006; 41:136-41.
15. Muñoz-Hoyos A, Rodríguez-Cabezas T, Molina-Carballo A, Martínez-Sempere JJ, Ruiz-Cosano C, Acuña-Castroviejo D. Melatonin concentration in the umbilical artery and vein in human preterm and term neonates and neonates with acute fetal distress. J Pineal Res 1992; 13:184-91.
16. Molina-Carballo A, Acuña-Castroviejo D, Rodríguez-Cabezas T, Muñoz-Hoyos A. Effects of febrile and epileptic convulsions on daily variations in plasma melatonin concentration in children. J Pineal Res 1994; 16:1-9.
17. Tarullo AR, Gunnar MR. Child maltreatment and the developing HPA axis. Horm Behav 2006; 50:632-9.
18. Blizzard RM, Bulatovic A. Psychosocial short stature: a syndrome with many variables. Baillieres Clin Endocrinol Metabol 1992; 6:687-712.
19. Powell GF. Nonorganic failure to thrive in infancy: an update on nutrition, behavior, and growth. J Am Coll Nutr 1988; 7:345-53.
20. Farrace S, Cenni P, Tuozzi G, Casagrande M, Barbarito B, Peri A. Endocrine and psychophysiological aspects of human adaptation to the extreme. Physiol Beh 1999; 66:613-20.
21. Cohrs S, Pohlmann K, Guan Z, et al. Quetiapine reduces nocturnal urinary cortisol excretion in healthy subjects. Psychopharmacology (Berl) 2004; 174:414-20.
22. Wetterberg L, Beck-Friis J, Aperia B, Petterson V. Melatonin/cortisol ratio in depression. Lancet 1979; 2:1361.
23. Wahlund B. Melatonin in psychiatric disorders - subtyping affective disorder. Biol Signals Recept 1999; 8:120-5.
24. Lewis M, Ramsay DS. Developmental change in infants' responses to stress. Child Dev 1995; 66:657-70.
25. Dahl RE, Siegel SF, Williamson DE, et al. Corticotropin releasing hormone stimulation test and nocturnal cortisol levels in normal children. Pediatr Res 1992; 32:64-8.
26. Khan R, Daya S, Potgieter B. Evidence for a modulation of the stress response by the pineal gland. Experientia 1990; 46:861-2.
27. Turner-Cobb JM. Psychological and stress hormone correlates in early life: a key to HPA-axis dysregulation and normalisation. Stress 2005; 8:47-57.
28. King S, Laplante DP. The effects of prenatal maternal stress on children's cognitive development: Project Ice Storm. Stress 2005; 8:35-45.
29. Glod CA, Teicher MH, Hartman CR, Harakal T. Increased nocturnal activity and impaired sleep maintenance in abused children. J Am Acad Child Adolesc Psychiat 1997; 36:1236-43.
30. Kirby AW, Clayton M, Rivera P, Comperatore CA. Melatonin and the reduction or alleviation of stress. J Pineal Res 1999; 27:78-85.
31. Kopp C, Vogel E, Rettori MC, Delagrange P, Misslin R. The effects of melatonin on the behavioural disturbances induced by chronic mild stress in C3H/He mice. Behav Pharmacol 1999; 10:73-83.
32. Naranjo-Rodriguez EB, Osornio AO, Hernandez-Avitia E, Mendoza-Fernandez V, Escobar A. Anxiolytic-like actions of melatonin, 5-metoxytryptophol, 5-hydroxytryptophol and benzodiazepines on a conflict procedure. Prog Neuropsychopharmacol Biol Psychiatry 2000; 24: 117-29.
33. Brotto LA, Gorzalka BB, LaMarre AK. Melatonin protects against the effects of chronic stress on sexual behaviour in male rats. Neuroreport 2001; 12: 3465-9.