Chapter 88

Biochemistry, Circannual and Circadian Rhythms, Endocrinology, and Immunology of Cluster Headaches

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BIOCHEMISTRY

Histamine and Mast Cells

Horton (64) suggested that cluster headache (CH) was associated with an unusual histamine sensitivity. The histamine levels have been reported to be higher during attacks than between attacks in whole blood (3) but not in urine (136). Increased numbers of mast cells, the main source of histamine, have been found in skin biopsy specimens from the painful temporal region both during and between cluster periods (5,127), but also from the pain-free side compared with control individuals. The mast cells were typically localized perivascularly, but in CH they were also found near cutaneous nerves (94). Increased degranulation, possibly as the response of an axon reflex, during and between cluster periods (5), a few hours after cluster attacks (68), and between attacks (29), as well as a normal degree of degranulation (20,73), has been observed. In basophils from cluster patients, increased degranulation after nitroglycerin challenge *in vitro* has been shown, a response that was normalized after incubation with lithium solution (129).

Prostaglandins and Leukotrienes

Prostaglandins, leukotrienes, and other eicosanoids have been proposed as possible mediators in the pathogenesis of vascular headache. Prostaglandins are local hormones that are produced at their site of action and have vasoactive properties. Although a tendency toward higher serum levels of prostaglandin E2-like substances during cluster attacks compared with basal values has been shown (117), which might be consistent with vasodilation, nonsteroidal antiinflammatory drugs generally have no therapeutic effect in CH and prostaglandins are not believed to be of primary pathogenetic importance. The leukotriene B4 (LTB4) has the ability to induce hyperalgesia and to enhance vascular permeability, but leukotrienes also have been reported to reduce nociceptive responses to bradykinins (133). During attacks, increased plasma levels of LTB4 analyzed by radioimmunoassay have been reported compared with the pain-free state (134), but with high-performance liquid chromatography the concentration of leukotrienes in cluster patients during and between attacks was below the detection limit of the method (77). During remission, the induced release of LTB4 and LTC4 from circulating basophils was significantly reduced (104) in CH patients compared with healthy controls.

Opioids

Plasma, cerebrospinal fluid (CSF), and platelet concentrations of metenkephalinlike (MET) and β-endorphin–like activities have been studied in patients during different phases of CH and in controls. CSF MET has been reported to be lower during (2,59,60) and between (60) attacks of CH or not to differ (149) compared with control patients. A decrease of CSF MET during attacks may reflect altered antinociceptive functions in CH. Low levels of CSF MET are probably not caused by increased degradation because the activities of enkephalinase and angiotensin-converting enzyme (ACE) in CSF at the lumbar level were not altered in CH patients compared with controls (137). In chronic CH, plasma MET levels have been found to be lower during and after attacks than before the attack (111). In episodic cases higher levels of MET in plasma have been reported during attacks (43,60), and higher (60) or normal levels were found between attacks and during remission (43). The increase in plasma MET during pain may be secondary

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to sympathetic activation because MET in circulation is derived mainly from the adrenals, where it is stored with catecholamines. It also may be derived from neutrophils, in which a decrease of MET has been observed parallel to the increase in plasma (44).

As regards β-endorphin concentrations in CSF at the lumbar level, no alterations have been found in samples obtained during and between attacks of CH (60). In plasma, β-endorphin has been reported to be increased during attacks (4) or normal (60) but without a normal circadian rhythmicity in several subjects (48,115). Beta-endorphin in plasma is released mainly from the pituitary gland, where it is synthesized from the same precursor as adrenocorticotropin (ACTH) and β-lipotropin. Lymphocytic β-endorphin is reviewed later in this chapter.

Nociceptin is an opioid neuropeptide with algesic and analgesic properties depending on the site of action. Circulating levels were lowered during cluster periods compared to healthy controls. Lower nociceptin levels were speculated to result in defective regulation of the trigeminal ganglion and insufficient protection against attacks (36).

Neuropeptides, Proteins

Much attention has been focused on the innervation of the cranial vasculature (see Chapter 9). In short, the large intra- and extracerebral vessels including the venous sinuses are supplied by the trigeminal nerve marked by calcitonin gene–related peptide (CGRP), substance P (SP), and neurokinin A (NKA); sympathetic nerve fibers marked by norepinephrine (NE), neuropeptide Y (NPY), and adenosine triphosphate (ATP); and parasympathetic nerve fibers marked by acetylcholine, vasointestinal peptide (VIP), and nitric oxide. The primary afferents of the trigeminal nerve not only transmit nociceptive information but also have efferent properties with the ability to dilate blood vessels, cause extravasation, and release from mast cells.

Local release of neuropeptides from the ipsilateral external jugular vein has been studied in CH. During spontaneous CH attacks CGRP and VIP levels were elevated, whereas there were no changes in NPY or substance P compared with a matched control group (30,56). The CGRP concentrations decreased to normal values within 15 minutes after administration of sumatriptan and oxygen as treatment but remained elevated after pethidine, although there was pain relief. The results indicate that sumatriptan, oxygen, and pethidine relieve pain by different mechanisms. The plasma levels of CGRP also have been reported to be elevated between cluster attacks and to be further increased during nitroglycerin-induced attacks but not by nitroglycerin per se (Fig. 88-1) (39,40) (see also Chapter 95).

Substance P can induce pain when combined with bradykinin, serotonin, or prostaglandins that lower the pain threshold. Activation of substance P fibers of trigeminal or facial nerve origin might explain the pain and the autonomous signs of CH and would be expected to cause an increase of CSF substance P levels, but in samples of spinal fluid from the lumbar level the substance P concentration has been reported to be normal both during (60,135) and between (53,60) attacks. In plasma, substance P is reported to be decreased and its degrading enzyme enkephalinase to be increased during cluster attacks (135). In saliva, CGRP, VIP, and substance P concentrations have been found to be higher during attacks than between attacks, and CGRP and VIP were higher both during and between attacks compared with controls (120).

Endothelin-1 in plasma have been reported to increase during attacks compared to between attacks but with no significant changes of arterial blood pressure (49).

Somatostatin is an inhibitory transmitter peptide found in sympathetic ganglia and in some primary afferent neurons, where it may inhibit substance P release from the C fibers. Somatostatin concentrations in plasma have been shown to be lower during (135) and between (14,135)

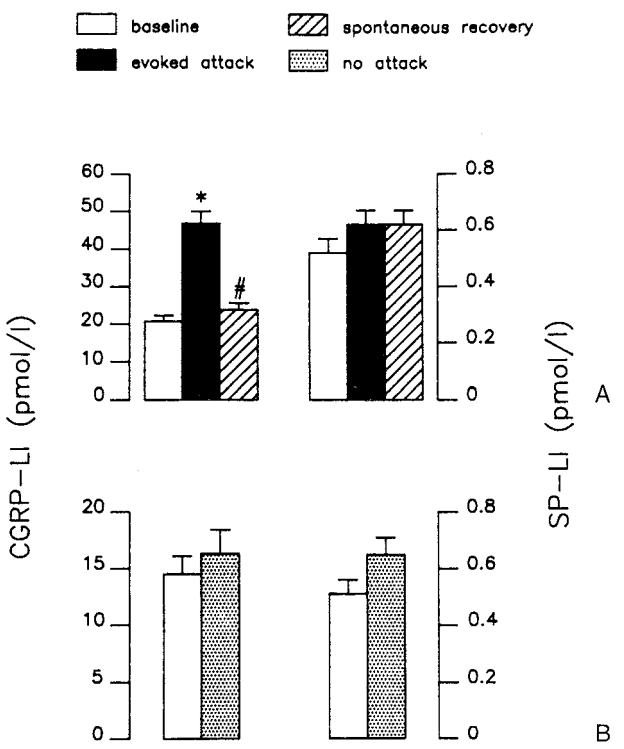

FIGURE 88-1. **A:** CGRP-Like and SP-Like measured in plasma from the external jugular vein of 12 cluster patients in an active period before nitroglycerin (*open columns*), at the peak of the provoked headache (*solid columns*), and after spontaneous remission (*dashed columns*). **B:** CGRP-Like and SP-Like measured in plasma from the external jugular vein of 12 cluster patients in remission period before (*open columns*) and 40 minutes after (*dotted columns*) nitroglycerin. The columns denote the means ± standard error of mean. Statistical significance (Duncan's test): $*p < 0.01$ versus values before nitroglycerin and $\#p < 0.01$ versus values at the peak of the attack. (Reprinted with permission from ref. 40).

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cluster attacks as well as during remission (14) compared with controls. An injection of somatostatin appeared to relieve CH pain (135). Somatostatin has venoconstrictive properties (14), which may be of importance for its therapeutic effect. Due to rapid tachyphylaxis, somatostatin is not likely to become a choice for CH treatment.

In CH there is an increased morbidity from peptic ulcer and a marked increase of gastric acid secretion during the attack phase. Gastrin levels in plasma have been studied in men between and during cluster periods. When compared with healthy controls, the gastrin levels were significantly higher in CH patients free of peptic ulcer symptoms (118).

Vasopressin has been shown to increase during CH attacks without any concomitant variation of plasma osmolality (47). Pain is one of several inducers of vasopressin secretion via catecholaminergic afferents from the brainstem to the hypothalamus. Serotonin also may trigger vasopressin release. As a potent vasoconstrictor, the increase of vasopressin has been suggested to counteract the vasodilatation associated with the attack (47).

Involvement of Gi proteins in the modulation of pain is widely established and Gi inactivation may produce hyperalgesia and insensitivity to analgesic treatment. Gi proteins inhibit adenylate cyclase and modulate several K^+ and Ca^{2+} channels. In lymphocytes from cluster headache and migraine patients but not in other pain syndromes, a hypofunctionality of Gi proteins has been demonstrated by a reduced capability to inhibit forskolin-stimulated adenylate cyclase activity (52).

Monoamines

Spontaneous cluster attacks often seem to be preceded by a shift of vegetative tone, indicative of increased parasympathetic or decreased sympathetic activity as during relaxation after meals or the working day or in relation to rapid eye movement (REM) sleep. Physical exercise has been shown to reduce the pain of induced attacks, and NE infusion also appeared to alleviate the pain (34). Nitroglycerin-induced attacks seemed to occur after an initial increase of plasma NE had reversed to basal values (66). A further increase in NE occurred during both spontaneous and induced attacks, maybe as a response to vasodilation. Because posture affects NE levels, the evaluation of results during attacks may be difficult (95). When examined before and after 5 minutes of standing, plasma concentrations of NE and epinephrine did not differ between patients in remission and controls, indicating normal postural responses (65). During cluster periods, contradictory results with both increased plasma concentrations of conjugated NE and epinephrine in plasma (93) and decreased concentrations of NE in the morning and at night in comparison with healthy controls were reported (141). The same researchers also found lowered CSF concentrations of vanillyl mandelic acid, homovanillic acid, 5-hydroxyindoleacetic acid, and NE in the active period (141).

Monoamine oxidase (MAO) activity in platelets has been documented in several studies to be lowered during and between cluster periods both in men and women, and when smoking habits are considered, but with no further decrease during cluster attacks (112). The decrease in MAO activity was explained by fewer enzyme molecules (11,142) and lowered V_{max} (capacity) with no change in K_m (affinity) (147). The enzyme is also more thermostable in CH patients than in controls (147). Because platelet MAO is of the B type, it does not metabolize NE, epinephrine, or serotonin (5-HT), but it may affect these amines indirectly by its decreased ability to catabolize dopamine, tyramine, and other trace amines. MAO is localized in the outer membrane of the mitochondria.

Succinate dehydrogenase (SDH), another membrane-bound mitochondrial enzyme, and the cytoplasmatic enzyme phenolsulphotransferase M (PST M) were analyzed in the same platelets as MAO (96). The SDH activity was significantly lowered in CH, whereas the PST M activity did not differ between patients and healthy controls. In the lowest range of MAO there was an inverse correlation between the activities of MAO and PST M, which was interpreted as a possible control mechanism compensating for a deficiency of one enzyme by another enzyme acting on the same substrates, for instance, dopamine and tyramine. Other platelet functions such as release of β-thromboglobulin and platelet factor 4 were decreased during CH attacks, contrasting with migraine, where increased release is known to occur during headache (22). The fibrinogen-binding properties of platelets from episodic and chronic cluster patients did not differ from those in controls (72). As regards 5-HT uptake into platelets, lowered V_{max} and lowered K_m (146) or no difference (58) in the kinetic parameters were reported in CH patients compared with controls. Factors such as time of the year (101,146) and medication may explain some differences in the results. Nitric oxide functions were suggested to be involved (22).

Nitric Oxide

The possibility to induce cluster headache attacks during the active period by glyceryltrinitrate, which is an exogenous NO-donor and histamine, which induces NO release from vascular endothelium suggested that NO is a key mediator in cluster headache. Excess NO production or increased activation of NO-ergic pathways might be involved. One study (17) showed that the increase of the NO metabolite nitrite after glycerylnitrate stimulation did not differ between patients during an induced cluster headache attack and healthy controls, and that basal levels of plasma nitrite did not differ between patients and controls. Neither did plasma levels of L-citrulline, a marker of

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endogenous NO-production, differ between patients and controls in the basal state. This was judged not to support hyperactivity of the L-arginine-NO pathway in cluster headache. In contrast increased plasma levels of nitrite were reported both in remission and cluster period between attacks as compared to controls. The discrepancies may be due differences in method and numbers of patients (21).

Trace Amines

The hypothesis that trace amines such as tyramine, octopamine, and synephrine, closely related chemically to classic biogenic amines, may contribute to the pathogenesis of primary headaches was proposed several decades ago. Trace amines displace biogenic amines from their storage vesicles and have the capability to act as false neurotransmitters. However, despite evidence showing that levels of trace amines in rodent brains are elevated during inhibition of MAO enzymes (MAO-A and MAO-B) or after selective deletion of MAO genes, to date there is no direct evidence supporting the involvement of trace amines in primary headaches.

Recently, however, G-protein-coupled receptors with high affinity for trace amines have been described in rodents and humans. These receptors, called trace amine receptors (TARs), are distinct from the classic biogenic amine receptors and are found in various tissues and organs, including specific brain areas such as the amygdala, hypothalamus, and locus ceruleus. In addition, effects of trace amines, in particular octopamine, on mammalian $\alpha 2$ and $\beta 2$ adrenergic receptors have emerged. All this opens the possibility that one or more trace amines may in humans behave as neurotransmitters or neuromodulators capable of exerting effects independently or in concert with classic biogenic amines.

A sensitive high performance liquid chromatography (HPLC) method for assessment of trace amines in human plasma and platelets was devised (23). Levels of tyramine, octopamine, and synephrine were, in comparison with healthy control subjects, assessed in patients experiencing CH during both active and remission periods.

In CH, the observation that significantly increased levels of the evaluated trace amines occur during both the remission and the active phases raises the hypothesis that such alterations may reflect the ongoing sympathetic dysfunction. Trace amines are known to be synthesized and stored within the autonomic nervous system. Thus, one possibility is that the increased plasmatic trace amine levels found in CH patients may reflect an increase of tyrosine decarboxylase activity or inhibition of tyrosine hydroxylase (TH) enzyme activity. In support of this hypothesis is the evidence showing that NE, the major product of TH activity, is decreased in platelets in all phases of CH and in plasma and CSF only in the active phase. In addition, the TH enzyme within the autonomic nervous system is the major source of NE in plasma. Another possibility is that the abnormal trace amine levels in CH may reflect hypothalamic dysfunctions. The hypothalamus and locus ceruleus contain, in humans, the highest level of octopamine, and these areas are connected with the autonomic system. Further, low prolactin levels in CH patients in all phases of the disease and after challenge with TRH have been reported. Although these findings may reflect dopaminergic hyperactivity, no evidence has been provided in support of this possibility; an alternative explanation may be an increased level of octopamine turnover in the hypothalamus. Octopamine, in fact, reduces prolactin secretion from lactotrophic cells via nondopaminergic receptors. In addition, hypothalamic abnormalities play a major role in the pathogenesis of CH. A study with positron emission tomography (PET) has demonstrated that regional cerebral blood flow, an index of synaptic activity, is increased during nitroglycerin-induced CH attacks in the posterior area of the hypothalamus (107). The same group, using voxel-based morphometry magnetic resonance imaging (MRI) analysis, has shown an enlarged volume of the gray matter in the same area (108). A new effective treatment, based on stereotactic stimulation of posterior hypothalamus in patients with intractable chronic CH, also supports a hypothalamic involvement (50). Interestingly, TAR-1 mRNA is reported to be expressed in the hypothalamus as well as in the ventral tegmental area, locus ceruleus, and dorsal raphe nucleus, structures that, among other functions, govern the pain threshold.

Recently, PET data of spontaneous CH attacks were published, revealing an activation pattern comparable with that observed in nitrate-induced CH (138).

This emphasizes that CH is not a primarily vascular disorder and that primary headache syndromes can be distinguished on a functional neuroanatomic basis by areas of activation specific to the clinical presentation.

Membranes and Phospholipids

Because a number of membrane-bound functions such as MAO-B, SDH, and PST M enzyme activities and 5-HT uptake in platelets are altered in CH as compared with healthy individuals, it is of interest to examine membrane composition and membrane transduction properties. Therefore, erythrocyte choline concentrations were measured in erythrocytes from CH patients and found to be depressed both during and between cluster periods to values about 50% of those found in controls (26), probably reflecting an abnormality in phospholipid metabolism. Thus, the phosphatidylcholine (PC) content of erythrocyte membranes, from which choline is derived, is increased, suggesting that the decreased concentration of choline could be explained by decreased PC turnover (28). Lithium treatment is known to increase the choline content of erythrocytes. Accordingly, a 12-week course of lithium treatment

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normalized the choline content in CH (26), which may be of importance for the prophylactic effect of lithium in CH. Altered receptor-mediated membrane transduction function involving the adenylate cyclase and the polyphosphoinositide (PPI) systems also have been demonstrated in CH (27). Adenylate cyclase, which is linked to surface receptors, is dependent on phospholipid constituents of the membranes for its activity. Accordingly, a significantly lower increase of the second messenger cyclic adenosine monophosphate has been shown in lymphocytes from CH patients than from controls after *in vitro* stimulation of high-affinity prostaglandin receptors, and a similar trend was shown after stimulation of β-adrenergic receptors (28). The PPI system in platelets stimulated with thrombin has been shown to have enhanced activity in untreated patients and normal activity in lithium-treated patients as compared with controls (27). It has been suggested that the prophylactic effect of lithium could be related to dampening of the activated PPI system to balance the reduced adenylate cyclase activity.

Further alterations in phospholipid metabolism involve a decreased ability to incorporate 1-14C-arachidonic acid and 1-14C-oleic acid into phosphatidylcholine and an increased ability to incorporate these fatty acids into phosphatidylserine and phosphatidylethanolamine, respectively (45,46). This also may affect membrane function because phosphatidylserine is required to bind the cytoplasmatic enzyme protein kinase C to the membrane during transmembrane signaling. Phosphatidylserine is also a source for polyunsaturated fatty acids in the synthesis of prostaglandins and leukotrienes.

Phosphorus magnetic resonance spectroscopy (31P-MRS) is a noninvasive method by which it is possible to measure high-energy phosphates and the efficacy of ATP production. A defect of brain mitochondrial respiration has been shown in CH both during and after a CH period compared with matched healthy volunteers (98,110). It was also reported that deficient energy metabolism was associated with low free magnesium in the occipital lobes in cluster headache and migraine (99). 31P-MRS of resting gastrocnemius muscle did not differ between patients and healthy controls, but after exercise phosphocreatine recovery was abnormally slow in the patients. The mechanism responsible for the multisystemic mitochondrial impairment is not known. It is suggested that the altered energy metabolism might render the patients more susceptible to metabolic demands during stressful conditions.

Excitatory Amino Acids

Platelets have been studied because they take up glutamate and aspartate by an energy-dependent mechanism similar to that occurring in neurons. Glycine levels in platelets were significantly lower in CH patients than in healthy controls, whereas the levels of aspartate and glutamate did not differ (22), contrasting with the findings in migraine with aura, where the concentrations of all three amino acids were increased. There was no difference in glycine levels during and between cluster attacks.

CIRCANNUAL AND CIRCADIAN PERIODICITY OF CLUSTER HEADACHE

The remitting course was clearly described by Ekbom (32), and a seasonal variation was noted by Horton (64). In certain patients cluster periods recur at regular intervals or fixed seasons, a pattern that suggests a relationship to environmental or internal factors such as day length and light intensity (76) or changes in stress (psychologic and physical) or activity, which can affect autonomous tone, including vascular regulation. The occurrences of the pain attacks after 1 to 2 hours of sleep, in the early morning, during relaxation periods (102), or with clockwise regularity (33) for at least part of the period are well-known phenomena. They suggest a central disturbance of circadian rhythm regulation affecting homeostasis of vascular and autonomic tone, nociception, and the synchronization of internal and environmental temporal clues. According to this paradigm, a cluster period would occur when there is desynchronization of these rhythms and would last as long as the time needed for resynchronization. An alteration in the circadian rhythm of plasma melatonin in CH indicates desynchronization of biologic rhythms. Melatonin, the main product of the pineal gland, is a marker of the circadian system. Its endogenous circadian secretory rhythm is driven by an oscillator in the hypothalamic suprachiasmatic nuclei, which are entrained to temporal variations of illumination via a retinohypothalamic norepinephric pathway. In humans, plasma melatonin levels are high at night and low during the day.

Reduced 24-hour plasma levels of melatonin, as well as phase shifts (advanced or delayed) in melatonin peaks, are observed during the cluster period (15,85–87,145). Stress increases melatonin secretion (148), and because the melatonin levels do not correlate with duration of illness, duration of headache in course, time since last attack, or attack frequency in CH (87), it is unlikely that lowered melatonin levels are due only to pain-induced stress (82).

In a multicenter study, urine melatonin was examined every month for up to 14 months in episodic CH patients (Fig. 88-2) (143); the 12-month mean levels of urinary melatonin were significantly lower in patients than controls, and no clear difference was found between cluster and remission periods (143). Similarly, nocturnal urinary excretion of 6-sulphatoxymelatonin, the chief metabolite of melatonin, is reduced in both illness phases (88). In some CH patients in the remission period, the circadian melatonin secretion is disrupted (typical rhythm lost). A similar melatonin disruption could also be present during the cluster period, but reduced melatonin levels would mask this. Reduced metabolism and

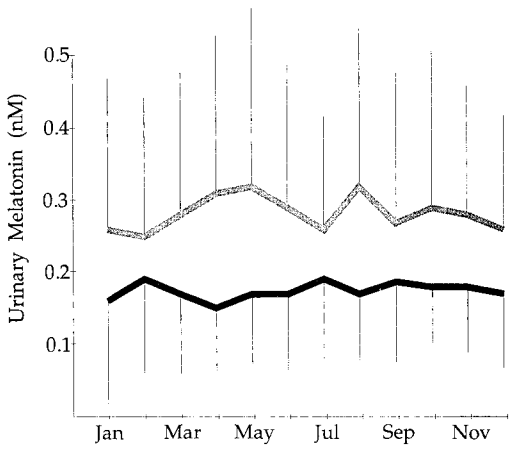

FIGURE 88-2. Chronogram of urinary melatonin concentrations covering 12 months in 29 episodic cluster headache patients (■) and in 29 healthy controls (⊞) matched to sex, nationality, and age. The effect of disease was significant ($p < 0.05$) but not the effect of time (Analysis of variance with repeated measurements; values are means ± standard deviation). (Reprinted with permission from ref. 143).

availability of serotonin for melatonin synthesis have been invoked to explain the reduced melatonin in cluster periods (145).

Reduced levels of melatonin could play a role in the pathophysiology of CH via several mechanisms: modulation of γ-aminobutyric acid (GABA) synapses in the central nervous system (7); modulation of the cellular distribution of calcium ions through binding of 3H-melatonin to calmodulin, thereby affecting the circadian variation of cell activity (6); involvement of melatonin receptors present in the main cerebral arteries (139); modulation of 5-HT2 receptor–mediated neurotransmission (31) (a property shared by the CH prophylactic methysergide); and inhibition of synthesis of prostaglandin E2 (78), involved in activating sterile perivascular inflammation in the trigeminovascular system. Oral melatonin administration has shown some efficacy as a CH prophylactic (83,84).

Serotonin, a precursor of melatonin, has a circannual variation with respect to platelet uptake (101). Plasma levels of NE and epinephrine show a circadian variation, with peak levels in the morning; this is in part a response to posture and sleep but probably also to a circadian oscillator (95). In healthy individuals, there is also a circadian rhythm of basal vascular tone due in part or entirely to increased sympathetic vasoconstrictor activity in the morning (123). This has not been studied in CH. Circadian variation in sensitivity to experimentally induced pain in the pericranial muscles (57) as well as of the flexion reflex (R III threshold) has been shown in healthy individuals. The R III threshold is lowest in the early morning and highest around midnight (132), but in CH during cluster periods the spontaneous physiologic variation of this threshold is lost, and by cosinor analysis there is no significant circadian rhythm (116). In addition, there is a lack of circadian rhythm of β-endorphin and β-lipotropin levels in blood in CH patients. These phenomena may support the hypothesis of cyclical failure of pain control in CH (114). In chronic CH there is no circadian rhythm of oral temperature or systolic and diastolic blood pressure, and in episodic cases a phase delay of these three variables is reported (116).

ENDOCRINOLOGY AND HORMONAL RHYTHMS

The Hypothalamic–Pituitary–Adrenal Axis: ACTH, Cortisol, β-endorphin, and β-lipotropin

The circadian production of cortisol is altered during cluster periods and in chronic CH (41,145). Moreover, a marked shift in the morning peak (acrophase) of the hormone characterizes these patients (15,114). Similar alterations are found in patients undergoing lithium treatment (16). Hyperactivity of the hypothalamic–pituitary–adrenal (HPA) axis in cluster periods is indicated by the finding that 24-hour cortisol production and morning levels are increased (15,86,87,141,145). Increased morning plasma cortisol is also seen in other head pain conditions (81,140). Morning cortisol levels are also high during the remission phase of CH (89), indicating that HPA axis overactivity is not simply a stress response. The reduced response of cortisol to the ovine corticotropin-releasing hormone test, observed in both remission and cluster periods, strongly suggests a condition of HPA axis hyperactivity (downregulation of the adrenal corticotrophic cells) that is not related to pain stress (89). On the other hand, blood cortisol is suppressed normally by the dexamethasone suppression test in both phases of CH (51,85), suggesting that the feedback mechanisms regulating blood cortisol are normal. A reduced response of blood cortisol and ACTH to the insulin-induced hypoglycemia test is reported both in cluster and in remission periods (89), suggesting that the hypothalamus may be hyporesponsive to hypoglycemia; the finding of reduced autonomic NE response to this test supports this (89). The finding of a reduced cortisol response to the 5-HT1A/2C agonist m-chlorophenylpiperazine in both CH phases (79,80) indicates that the reduced responsiveness of the HPA axis in CH is mainly due to a hypothalamic 5-HTergic dysfunction. In addition, investigation of the pro-opiomelanocortin-related peptides β-endorphin and β-lipotropin has revealed anomalies in their circadian production during CH (48,114).

Prolactin

The diurnal rhythm of prolactin has been reported to be normal (15,41) or altered (42,126) in the cluster period.

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That prolactin regulatory mechanisms are in fact compromised in CH is indicated by the finding that prolactin production over 24 hours is reduced in both phases of the illness (144). This reduction does not seem to be secondary either to sleep alterations or to the pain episodes. Altered central dopaminergic regulation of prolactin in CH is indicated by a smaller than normal increase in prolactin levels following stimulation with the D2 antagonist metoclopramide (70). A response of this type indicates that the lactotrophic cells of the hypophysis are downregulated. Downregulation by thyrotropin-releasing hormone (TRH) also could explain the reduced response of prolactin to the TRH test found in female patients in both phases of CH and in chronic CH (144). The lack of prolactin increase after morphine administration (9) indicates that opioid regulation of the hormone also may be compromised in CH and further suggests that the activation threshold for the mechanisms concerned with response to stress stimuli may be altered in CH, as is also indicated by the blunted cortisol and ACTH responses to the insulin test (89). The increased prolactin response to the 5-HT1A/2C agonist m-chlorophenylpiperazine only during cluster periods is probably due to 5-HT1A hypersensitivity as indicated by the normal prolactin response both to the 5-HT2C receptor antagonist cyproheptadine (71) and the 5-HT3 antagonist quipazine (125). This response is not specific and is probably a pain-induced stress phenomenon (87,93).

Testosterone and Sex Hormones

Several studies have reported reduced morning levels of testosterone in CH (38,69,71,74,113,119,128,144). Lowered production over 24 hours (38) and a phase shift of the morning peak (38,144) also have been described. These changes do not seem to stem from luteinizing hormone (LH) changes (144), although fewer LH peaks are seen in CH patients (109). Nor can they be explained by changes in testosterone metabolism or by an altered sleep–wake cycle, although the latter is found in some patients with nocturnal pain crises (144). Possibly, lowered testosterone levels in CH are secondary to the increased plasma cortisol levels (144). Estrogens, progesterone, and their metabolites are normal in CH (113,144).

Luteinizing Hormone and Follicle-Stimulating Hormone

There is no circadian rhythm of LH or follicle-stimulating hormone (FSH) in healthy controls or in CH patients; serum levels are also normal (144), but fewer and more prolonged LH peaks over 24 hours are observed in the cluster period (109). The regulation of gonadotropin production has been investigated by means of various tests. A reduced response of LH to luteinizing hormone–releasing hormone (LHRH) challenge, associated with increased FSH response, has been observed in the cluster period and in chronic CH patients (113). This type of response is also observed in prepubescent and postmenopausal subjects, in whom central control of sex hormones (as occurs in fertile adults) is lacking. However, other studies have not confirmed altered secretion of LH in response to challenge with LHRH (37). Opioid and serotoninergic control of hypophyseal gonadotropins appears normal in CH patients when probed by the naloxone (37) and quipazine (125) tests.

Growth Hormone

The circadian rhythm of growth hormone (GH) also has been found to be altered in cluster periods (15). Pain attacks and GH levels are not clearly correlated (8,15), suggesting that stress is not implicated in the genesis of GH alterations. Challenge with metoclopramide (a dopamine antagonist) (61), but not with L-dopa (a dopamine agonist) (8), induces an excessive GH response in CH patients. This response appears to be specific to CH because in other pain conditions metoclopramide challenge produces the same response as in healthy subjects (70). The finding that the GH response to insulin-induced hypoglycemia is normal in CH (8) contrasts with the reduced responses of ACTH, cortisol, and NE to this challenge reported by others (89). This discrepancy is probably due to different activation thresholds (to hypoglycemia) of the two hypothalamic systems controlling the release of GH and ACTH. Serotoninergic regulation of GH is normal on challenge with quipazine (125) and cyproheptadine (71).

Thyrotropin-Stimulating Hormone

Basal thyrotropin-stimulating hormone (TSH) levels are normal in CH (10,144), but a reduced TSH response to the TRH test characterizes patients during the cluster period (10,90). This alteration persists even when prophylactic treatment completely controls the pain crises (12). A blunted TSH response to the TRH test is also found in patients suffering from endogenous depression (100), again pointing to a central origin for these abnormalities in CH.

IMMUNOLOGY

There is growing interest in the relationship between the central nervous system and the immune system. It is known, for instance, that lesions to certain brain areas, particularly the hypothalamus and hippocampus, can induce specific modifications in immune system activity, probably mediated by changes in autonomic outflow to lymphoid organs (18). Animal studies have shed light on the role played by the central nervous system in immune system modulation. For instance, an immune response can be

evoked by conditioning of behavior in the rat (1); thus, it is evident that changes in the immune system may reflect altered cerebral signaling to lymphoid organs.

Lymphocyte Subpopulations

Natural killer and monocyte cell populations are increased in the cluster period compared with the remission phase (54,61). However, stress is known to influence immune system parameters (122), and to evaluate the influence of pain on these changes, immune parameters in CH patients and low back pain patients have been compared. Increased monocytes and reduced CD3+ (all T-lymphocytes) and CD4+ (T-helper) cells were observed both in cluster period and in low back pain patients (13). Because the number of natural killer cells in cluster period patients was similar to that in controls, immune alteration in CH is partly secondary to pain-induced stress. However, monocyte and natural killer cell levels are also reduced during remission (whereas CD3 and CD4 are within the normal range) (13), and because these patients are not pain stressed, the possibility arises that the alterations are an expression of deranged central monoaminergic modulation to the immune system (13).

Cytokines

An increase in the lymphokine-activated killer (LAK) cell phenomenon induced *in vitro* by adding interleukin-2 has been described in both phases of CH (55). This is likely to be caused by an increase in the number of cells bearing the interleukin-2 receptor. The elevated soluble interleukin-2 receptors indicate T-cell activation and suggest immune activation during CH. Because interleukin-2 can activate the hypothalamus and stimulate the release of corticotropin-releasing factor (CRF), interleukin-2 could link a putative immunologic cause of CH with the observed hypothalamic activation (35). Interleukin-1α and interleukin-1 levels, on the other hand, are reported to be normal in CH (13). Interleukin-1β has been reported to be increased (105).

Immunoglobulins and Complement

Concentrations of circulating immune complexes and of immunoglobulins are normal in CH patients (150); similarly, anticardiolipin antibodies are absent and the venereal disease research laboratory reaction is negative in CH sufferers (63). Both the classical and the alternative activation pathways of the complement system are also normal (150). Sedimentation rate, C-reactive protein, electrophoresis of serum, von Willebrand's factor, antinuclear antibodies, rheumatic factor, cytoplasmic and perinuclear antineutrophil cytoplasmic antibodies were all within normal limits compared with remission and healthy controls indicating that there are no clinical or laboratory signs of systemic inflammation during the cluster period (121).

Human Leukocyte Antigen

It is known that histocompatibility antigens are implicated in the regulation of testosterone in mice (67), and studies on the human leukocyte antigen (HLA) system in CH were prompted by the finding that testosterone levels are reduced in CH patients (66). Increased levels of HLA A1 (19), B35, and DR5 (54) and reduced levels of A3 (103), B14 (54,103), and B21 (103) have been reported. However, none of these modifications has been corroborated by other researchers, and some researchers report different HLA alterations.

Lymphocyte β-endorphin

It was discovered only recently that β-endorphin is normally present in peripheral blood lymphocytes (62). The mRNA for the β-endorphin precursor proopiomelanocortin is also present, indicating that β-endorphin is produced endolymphocytically, a conclusion further supported by the observation that lymphocytes are unable to take up β-endorphin from the extracellular medium. Lymphocytic β-endorphin is modulated by serotonin, dopamine, and GABA in much the same way that these neurotransmitters regulate β-endorphin levels in the hypothalamus (97,106,130,117). It also has been noted that in certain brain pathologies, for example, schizophrenia, alterations in lymphocyte peptides are associated with closely analogous changes in CSF (124). These observations suggest that monitoring lymphocytic β-endorphin can provide indirect information on β-endorphin levels in the brain. Lymphocytic β-endorphin is markedly reduced in CH patients, both in the cluster period and in remission (92). This alteration is unrelated to the pain attacks and may indicate a diffuse opioid system derangement in this illness, but does not seem specific for CH (91).

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P1: KWW/KKL P2: KWW/HCN QC: KWW/FLX T1: KWW
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