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**Title**

**THE ROLE OF THE HYPOTHALAMO-PITUITARY AXIS IN HEADACHE**

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**Supervised by Professor Peter Goadsby**

**A thesis submitted in part fulfilment for the degree of Medical Doctorate (M.D) at the  
Headache Group, Institute of Neurology, Queen Square, University College London,  
2004**

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**For Ben**

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## **Abstract**

This thesis investigates the hypothesis that dysfunction of the hypothalamo-pituitary axis is important in headache in patients with pituitary disorders. The observation that patients with small functional pituitary tumours may suffer with severe headache syndromes, and the dramatic analgesic effect that may be seen with somatostatin analogues, are central to the thesis. In the first study, I showed that there was no correlation between pituitary volume or cavernous sinus invasion with headache in pituitary tumours. In this study, prolactinomas and growth hormone- secreting tumours were found to be associated with the highest degree of headache, suggesting that biochemical mechanisms may be more important than structural ones in the pathophysiology of pituitary tumour-associated headache. In the second study, the presence of potentially nociceptive peptides CGRP and substance P within pituitary tumours was investigated; there was no association between the presence of these peptides and headache. In the third study, the clinical characteristics of pituitary tumour-associated headache were investigated. The commonest presentation of headache was migraine. The rare primary headache, Short lasting Unilateral Neuralgiform headache attacks with Conjunctival injection and Tearing (SUNCT), was exclusively associated with acromegaly or prolactinoma. In the fourth study, the possibility that cessation of somatostatin infusion could be a useful non-vascular way of triggering headache was investigated; the headache induction was not reliable. In the final study, the potential use of octreotide in the management of primary headache was investigated. Octreotide was unhelpful for migraine, but was efficacious in the acute treatment of cluster headache. The findings suggest that functional disturbances in the hypothalamo-pituitary axis may have a pathophysiological role in some headache types.

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## **Publications arising from thesis**

### Papers

Octreotide is not effective in the acute treatment of migraine. Levy MJ, Matharu MS, Bholra R, Meeran K, Goadsby PJ. *Cephalalgia* 2004; 24

Subcutaneous octreotide is effective in the treatment of acute cluster headache: a double-blind placebo-controlled crossover study. Matharu MS, Levy MJ, Meeran K, Goadsby PJ. *Annals of Neurology* 2004 (in press)

Pituitary volume and headache; size is not important. Levy MJ, Matharu MS, Jager HR, Powell M, Goadsby PJ. *Archives of Neurology* 2004; 61(5): 721-5

Somatostatin-triggering of headache. Levy MJ, Matharu MS, Lightman SL, Goadsby PJ. *Pain* 2003; 102 (32): 237-242

Prolactinoma, dopamine agonists and headache. Levy MJ, Matharu MS, Goadsby PJ. *European Journal of Neurology* 2003; 10:1-5

Acromegaly; a unique human headache model. Levy MJ, Goadsby PJ, Meeran K. *Headache* 2003; 43 (7): 794-798

SUNCT syndrome secondary to prolactinoma. Matharu MS, Levy MJ, Merry RT, Goadsby PJ. *Journal of Neurology, Neurosurgery and Psychiatry* 2003; 74 (11): 1590-2

### Abstracts

Octreotide dependency: a cautionary tale. Levy MJ and Goadsby PJ (2004) *Endocrine Abstracts* 7; P110

Phenotypic characterisation of the headache associated with pituitary tumours. Levy MJ, Matharu MS, Powell M, Meeran K, Goadsby PJ (2003) *Cephalalgia* 23: 621. P2D6

Differential expression of CGRP and substance P in pituitary adenomas; in search of a nociceptive peptide. Levy MJ, Classey J, Maneesi S, Powell M, Meeran K, Goadsby PJ (2003) *Cephalalgia* 23: 687. P4M13

Comparative expression of NPY and Orexin B in the anterior pituitary tumours of patients suffering from headache. Classey JD, Levy MJ, Powell M, Grouzman E, Meeran K, Goadsby PJ (2003) *Cephalalgia* 23: 687. P4M12

Subcutaneous octreotide is effective in the treatment of acute cluster headache: a double-blind placebo-controlled crossover study. Matharu MS, Levy MJ, Meeran K, Goadsby PJ (2003) *Cephalalgia* 23: 687. 6.4



## **The role of the hypothalamo-pituitary axis in headache**

### **1.1 An introduction to headache**

There have been major advances in the scientific understanding of the pathophysiology of headache in recent years, both through clinical and animal research. One of the primary shifts in emphasis has been to move away from the view that headache is a 'vascular disorder' towards the view that it is a central nervous disorder (May and Goadsby, 1999). The trigeminal nucleus has been shown to be crucially involved in head pain and the classical human experiments performed by Wolff (1963) demonstrated that direct stimulation of the cerebral cortex and other central structures were not painful, whereas stimulation of areas innervated by the trigeminal nerve, such as the dura mater and cerebral blood vessels, produced pain.

Although the primary disturbance in headache syndromes is believed to be in the central nervous system, many patients report a vascular component to their symptoms (Drummond and Lance, 1983). This is believed to be a result of activation of the 'trigeminovascular' pathway, which describes an anatomical connection between trigeminal neurones and the cranial blood vessels. Stimulation of the trigeminal ganglion in the cat leads to an increase in vasoactive peptides such as substance P (SP) and calcitonin gene related peptide (CGRP; Goadsby et al., 1988) and stimulation of more specific pain-producing structures, such as the superior sagittal sinus, also results in the release of CGRP but not SP (Zagami et al., 1990). During migraine (Goadsby et al., 1990; Gallai et al. 1995), cluster headache (Goadsby et al., 1990; Fanciullacci et al., 1995) and other primary headache syndromes (Goadsby and Edvinsson, 1996), CGRP has been shown to be elevated, which strongly implicates a role for this peptide in the pathophysiology of headache. CGRP (Steel et al., 1992; Nakamura et al., 1998) and SP (Arita et al., 1994; Liu, 1995) have both been isolated in the pituitary gland, as will be discussed later in the introduction.

Headache is a common feature of pituitary tumours and is a major cause of morbidity in this group of patients (Suwanwela et al., 1994; Abe et al., 1998). Acromegaly and prolactinoma appear to be particularly associated with headache (Abe et al., 1998;

Ezzat et al., 1994; Couch, 1986). As well as the high prevalence of headache in pituitary tumours, rare headache syndromes have been described in association with small hormonally active lesions (Massiou et al., 2002; Ferrari et al., 1988). An impressive analgesic effect of somatostatin analogues on pituitary tumour-associated headache has been described in the absence of reduction tumour size (Sandler et al., 1987; Williams et al., 1987; Musolino et al., 1990). The mechanism of this analgesic action is unknown, although it has been suggested that the suppression of a nociceptive peptide may be involved (Williams et al., 1987).

The title of this thesis is 'The role of the hypothalamo-pituitary axis in headache'. It is hoped that the subject matter within this thesis will facilitate information exchange between the endocrinologist interested in pituitary disease, and the neurologist interested in headache. This introduction attempts to draw together the current data regarding the involvement of the hypothalamo-pituitary axis in primary headache syndromes, the clinical presentation and proposed mechanisms of pituitary tumour-associated headache, and the perceived role of somatostatin in headache.

The first part of the introduction is a clinical and pathophysiological review of headache, with particular emphasis on the involvement of the hypothalamo-pituitary axis. This section includes a series of case histories of patients with pituitary tumours presenting with headache that exemplify the important issues that were generated whilst planning the work. The individual cases are fully described in the Appendix. In the second part of the introduction, a general background to the subject of pituitary tumours is given, with particular emphasis on headache. This section reviews the previous clinical reports of pituitary tumour-associated headache and includes a discussion of the potential pathophysiological mechanisms involved. The final part of the introduction discusses the topic of somatostatin and headache, following which is a summary of the aims of the individual studies in this thesis.

## **1.2 Case Histories**

One of the impetuses for this project was a series of observations made by two independent groups (the Endocrine Unit, Charing Cross Hospital, London and the Headache Group, Institute of Neurology, London). The cases described in the Appendix exemplify the type of presentations that aroused interest in this subject, and

are summarised here.

Cases 1 and 2 (Appendix) describe two similar patients presenting with very small pituitary tumours with severe headache (Levy et al., 2003a). These cases suggest that factors other than tumour size may be important in pituitary tumour-associated headache. Both had microprolactinomas and presented with rare headache syndromes at the same time as the onset of menstrual disturbance and galactorrhoea. Case 1 had a diagnosis of short lasting unilateral neuralgiform headache attacks with conjunctival injection and tearing (SUNCT; Goadsby and Lipton, 1997; I.H.S code 3.3, Headache Classification Committee of The International Headache Society, 2004) and case 2 had symptoms suggestive of hemicrania continua (Pareja et al., 2001; I.H.S code 4.7, Headache Classification Committee of The International Headache Society, 2004), the clinical features of which are discussed later in this introduction. The interesting aspect of these cases is the manner in which the administration of dopamine agonists significantly altered and exacerbated the headache phenotype without causing a change in the size of the lesions. The importance of size in pituitary tumour-associated headache is studied in Chapter 2.

Case 3 (Appendix) describes a patient with acromegaly who presented with severe unilateral headache (Levy et al., 2003b). The striking aspects of this case were the dramatic analgesic effect of octreotide and the unresponsiveness of the headache to lanreotide, despite adequate growth hormone suppression with both. The fact that lanreotide successfully reduced growth hormone but not headache suggests that octreotide possesses a specific analgesic property that is 'uncoupled' from growth hormone. The possibility that pituitary tumours might secrete a nociceptive peptide is investigated in Chapter 3.

Case 4 (Appendix) describes a 37 year-old man with a macroprolactinoma presenting with SUNCT (Matharu et al., 2003). The headache symptoms were ipsilateral to the side of cavernous sinus invasion and these symptoms completely resolved in response to dopamine agonist therapy. The clinical features of SUNCT and other rare headache syndromes are discussed later, and the range of headache phenotypes seen with pituitary tumours is investigated in Chapter 4.

Case 5 (Appendix) describes octreotide dependency (Levy and Goadsby, 2004), which has been previously reported (Popovic et al., 1988; May et al., 1994) although this is not widely recognised. The presence of rebound headache and tachyphylaxis in this patient indicates that overexposure to somatostatin can cause worsening headache. The potential usefulness of this observation for research purposes are studied in Chapter 5, and the potential role of octreotide as an abortive analgesic agent in primary headache is studied in Chapter 6.

In summary, pituitary tumours appear to give rise to a range of headache syndromes that may be caused by factors over and above simple structural mechanisms. The observation that headache can be alleviated or exacerbated by the administration of dopamine agonists or somatostatin analogues suggests that previously unexplained neuro-endocrine pathways may be important in the pathophysiology of certain headaches.

### **1.3 The hypothalamo-pituitary axis and headache**

In order to understand the significance of pituitary tumour-associated headache, it is important to have a clinical and pathophysiological perspective of headache in general. An important aspect of headache in clinical practice is the recognition that pain is only a small part of the presentation of most headache syndromes (Silberstein et al., 2002). Many of the non-painful clinical features associated with primary headache suggest that there are alterations in the hypothalamo-pituitary axis before, during and after a headache attack (Giffin et al., 2003). The following is a discussion of the different headache phenotypes mentioned in this thesis, with particular emphasis on the relevance of the hypothalamo-pituitary axis to each phenotype.

### **1.4 Migraine**

Migraine is a common clinical problem, having a one-year prevalence of approximately 17 % among women and 10 % among men (Steiner et al., 2003). Migraine has many facets other than pain in its clinical presentation, and this is well recognised by patients (Silberstein et al., 2002). To establish a diagnosis of migraine, as defined by the Headache Classification Committee of the International Headache Society (1988), having regard to its recent revision (2004), five attacks are needed, each lasting 4-72 hours and having two of the following four pain characteristics:

unilateral location, throbbing quality, moderate to severe intensity and aggravation with movement. In addition, the attacks must have at least one of the following: nausea or vomiting or photophobia and phonophobia. Functional imaging studies have suggested that the brain stem is activated during a migraine attack (Weiller et al., 1995; Bahra et al., 2001; Matharu et al., 2004) supporting the view that migraine is a central nervous system rather than a peripheral disorder. There are characteristic clinical features of migraine that implicate the hypothalamo-pituitary axis in the pathophysiology of migraine which are now discussed.

#### *1.4.1 Sex hormones and migraine*

Migraine is approximately twice as common in women (Stewart et al., 1994) and there is a known association between alterations in the female endocrine axis and migraine. Women commonly experience migraine peri-menstrually, which may be due to oestrogen withdrawal (Somerville, 1975; Silberstein, 2001). The role of fluctuating oestrogen levels in migraine is further evidenced by the observation that migraine attacks occur during the 'pill free interval' in women on the combined oral contraceptive pill (MacGregor, 1997) implying that oestrogen withdrawal is important in the pathophysiology of migraine. There are also a group of patients who experience oestrogen-induced migraine (Silberstein, 2000a) indicating that migraine may be exacerbated by both increases and decreases in oestradiol concentration. Migraine often improves during the second and third trimester of pregnancy when oestrogen and progesterone levels are persistently elevated, often exacerbating soon after delivery (Silberstein, 2000b). Migraine commonly worsens at the peri-menopause (Neri et al., 1993) further indicating the importance of fluctuations in oestradiol levels in relation to migraine. A well-recognised time for women to experience their first migraine attack is at menarche (Facchinetti et al., 2000), when there is much change in activity of the hypothalamo-pituitary axis (Styne, 1994).

#### *1.4.2 Premonitory symptoms*

Premonitory symptoms are probably under-recognised, partly due to their subtle nature (Santoro et al., 1990) and implicate hypothalamic disturbances in migraine. They should not be confused with aura, which occurs up to 60 minutes before the onset of pain, and occurs in 15% of migraineurs (Russell et al., 2002). Premonitory symptoms include increased yawning, tiredness, mood disturbances, polyuria,

polydipsia and altered food cravings (Drummond and Lance, 1984), strongly pointing towards hypothalamic disturbance (Kupfermann, 1985; Zurak, 1997). The incidence of premonitory symptoms ranges from 7-88% of migraine sufferers (Drummond and Lance, 1984; Waelkens, 1985; Rasmussen and Olesen, 1992) and these symptoms are highly predictive of attacks in affected patients (Giffin et al., 2003).

#### 1.4.3 Dopamine-prolactin axis

Premonitory symptoms may be due to alterations in dopaminergic pathways (Peroutka, 1997). Yawning may be a prominent premonitory symptom (Russell et al., 1996) and there is *in vivo* evidence that dopamine (D<sub>2</sub>) receptor activation can lead to experimentally-induced yawning (Mogilnicka and Klimek, 1977; Protais et al., 1983; Serra et al., 1986; Yamada et al., 1986). D<sub>2</sub> agonists may induce both yawning and headache in migraineurs (Blin et al., 1991; Del Bene et al., 1994). Migraine is less common in patients with Parkinson's disease than those without, and the frequency of migraine in Parkinsonian patients increases with administration of D<sub>2</sub> agonists (Sabatini et al., 1990). Migraineurs may have increased dopamine sensitivity (Cassidy et al., 2003) and abnormalities of D<sub>2</sub> receptors (Peroutka et al., 1997; Dichgans et al., 1998). There is clinical evidence that dopamine antagonists are helpful in the acute treatment of a migraine attack beyond their anti-emetic effects (Sharma et al., 2002; Silberstein et al., 2003).

Because of the intricate relationship between dopamine and prolactin (dopamine is 'prolactin inhibitory factor' when produced by the hypothalamus), there has been interest in the relationship between serum prolactin *per se* and migraine (Murialdo and Polleri, 1987). Peres et al. (2001) showed a reduction in serum prolactin in chronic migraine, whilst other groups have shown an elevation in serum prolactin in episodic migraine (Murialdo et al., 1986; Nattero et al., 1986). It is unclear whether this difference in findings is related to the episodicity of migraine. There is an increased prolactin response after pituitary stimulation in episodic migraineurs compared to healthy controls (Awaki et al., 1989) and an increased prolactin response to dopamine antagonists in the follicular phase of migraineurs (Murialdo et al., 1986). Conversely the inhibitory effect of nomifensine, a dopamine reuptake inhibitor, on prolactin secretion is dampened in migraineurs compared with controls (Murialdo et al., 1986). These findings may demonstrate an increased prolactin reserve in migraine

and suggest the existence of a dopaminergic supersensitivity of the lactotrophic postsynaptic D<sub>2</sub> receptors. A further link between dopamine and migraine is the observation that bromocriptine may exert an analgesic effect in primary migraine (Herzog, 1997) and in pituitary tumour-associated migraine (Hartman et al., 1995; Gabrielli et al., 2002).

#### *1.4.4 Migraine genetics*

One of the key features of migraine is its strong familial tendency. This was noted as far back as the eighteenth century (Tissot, 1790) and numerous subsequent studies have confirmed this. Familial studies support the familial aggregation of migraine (Merikangas, 1990; Merikangas, 1996) and twin studies also support the role of genetic factors (Merikangas, 1996). The observation that there is only an approximately 50% concordance in monozygotic twin studies suggests that the aetiology of migraine is multifactorial. It is unlikely that there is a single 'migraine gene' and abnormalities in the following genes have been linked to migraine: dopamine receptors (Peroutka et al., 1997), Notch 3 (Davous, 1998), mitochondrial genes (Klopstock et al., 1996), endothelin type A receptor (Tzourio et al., 2001), Xq24-28 linked gene (Nyholt et al., 2000) and the insulin receptor gene (McCarthy et al., 2001). There has been interest in the CACNA1A gene on chromosome 19p13 (Ophoff et al., 1996). This gene encodes the  $\alpha_{1A}$  subunit, which forms the calcium-selective pore of voltage gated P/Q-type calcium channels. CACNA1A mutations are associated with familial hemiplegic migraine, a rare autosomal dominant form of migraine with prolonged aura, hemiparesis and, in some families, cerebellar ataxia and atrophy. There has also been an association between familial hemiplegic migraine and the ATP1A2 gene on chromosome 1q23 (De Fusco et al., 2003; Vanmolkot et al., 2003). This gene encodes the  $\alpha_2$  subunit of the sodium-potassium pump, further implying that certain forms of migraine may be due to abnormalities in the function of ionic channels (so-called 'channelopathies').

In summary, there is evidence that patients with migraine have an inherited tendency to a condition that includes central neuroendocrine disturbances. This knowledge may be important in understanding the reasons why certain patients with functional pituitary tumours are particularly prone to headache.

## **1.5 Trigeminal Autonomic Cephalgias**

One of the observations made in the clinical cases described in the Appendix, supported by the literature (Ferrari, 1988; Massiou, 2002; Milos, 1996), is that cranial autonomic symptoms may be a feature of pituitary tumour-associated headache.

Cranial autonomic symptoms are a particularly prominent clinical feature of a group of primary headaches known as the trigeminal autonomic cephalgias, of which cluster headache is the most commonly described (Goadsby and Lipton, 1997). The cranial autonomic symptoms include ipsilateral parasympathetic activation (lacrimation, rhinorrhoea, nasal congestion, and eyelid oedema) and sympathetic hypofunction (ptosis and miosis). The mechanism of autonomic activation has been well described (Goadsby and Lipton, 1997). Stimulation of trigeminal efferent neurones can result in increased cranial autonomic outflow (May and Goadsby, 1999; Frese et al., 2003), known as the trigeminal-autonomic reflex. The trigeminal-autonomic reflex refers to a functional reflex between the trigeminal nucleus caudalis and the parasympathetic outflow from the superior salivatory nucleus.

### **1.5.1 Cluster Headache**

The tendency of cyclical/ seasonal expression in episodic cluster headache is strongly suggestive of hypothalamic involvement because the suprachiasmatic nucleus of the hypothalamus is believed to be the pivotal area of the brain controlling biological rhythms (Swaab et al., 1996). Functional imaging techniques, such as functional fMRI and Positron Emission Tomography (PET), along with evidence from biochemical investigation of patients with cluster headache, have shown that the hypothalamus plays a central role in the pathophysiology of this rare headache disorder. Therefore cluster headache gives numerous insights into the potential role of the hypothalamo-pituitary axis in headache.

#### *1.5.1.1 Clinical features of cluster headache*

A typical cluster attack is characterised by severe unilateral orbital/ supraorbital/ temporal pain lasting 15 to 180 minutes if untreated. Headache is usually associated with pronounced cranial autonomic features and the frequency of attacks varies from 1 every other day to 8 per day (Headache Classification Committee of the International Headache Society, 2004). Cluster headache may be episodic, occurring



in periods lasting 7 days to one year separated by pain free periods lasting 14 days. Cluster headache may also be chronic, occurring for more than one year without remission or remission lasting less than 14 days. In episodic cluster headache, a cluster period usually lasts between 2 weeks and 3 months. Unlike migraineurs (during a migraine attack patients are generally sedate and prefer to lie in a darkened room), patients with cluster headache are often extremely agitated and unable to lie down during an attack. Patients with cluster headache describe rocking, applying pressure to their head, banging their heads against walls, and other restless activities during an attack (Blau, 1993). This behavioural difference from migraine may not be just a reflection of the higher pain severity, but may be due to hypothalamic activation. It is known that hypothalamic centres are important for autonomic fight/flight responses (Palkovits, 1999) and aggressive behaviour (Panksepp, 1971; Kruk et al., 1983; Roeling et al., 1993) and this may explain the extreme agitation observed during a cluster attack.

#### *1.5.1.2 Biochemical abnormalities in cluster headache*

##### *Hypothalamo-Pituitary Adrenal Axis*

There is evidence that cortisol diurnal variation is lost in cluster headache, and this does not appear to be related to whether the attacks are nocturnal or during the day (Ferrari et al., 1983; Chazot et al., 1984; Facchinetti et al., 1986).

Twenty-four hour urinary cortisol production is increased in cluster headache during a bout compared with controls (Facchinetti et al., 1986; Waldenlind et al., 1987) implying activation of the adrenal axis. There is a reduced cortisol response to painful stimuli compared to healthy controls (Chazot et al., 1984; Waldenlind et al., 1987) as well as a reduced cortisol / ACTH response to CRH both in the cluster period and during remission (Leone et al., 1991), and reduced response to insulin-induced hypoglycaemia (Leone et al., 1991). The raised resting activity of the hypothalamo-adrenal axis in cluster headache suppresses normally in response to low-dose dexamethasone (Devoize et al., 1986; Frediani et al., 1988; Leone et al., 1992) suggesting that there is no autonomous secretion of cortisol in cluster headache.

### *Gonadal Axis*

Cluster headache is more common in males, with a male: female ratio of 7:1 in middle age (Ekbohm et al., 2002) although the ratio may be closer to 2.5:1 (Bahra et al., 2002; Torelli et al., 2003). The male predominance of cluster headache and the observation that it is very rare in puberty (Manzoni et al., 1991) suggests that the FSH/ LH/ testosterone axis may be important in cluster headache.

There is reduced basal LH (Kudrow, 1977; Klimek et al., 1984) and elevated FSH levels (Murialdo et al., 1989) in episodic and chronic cluster headache, although groups have also reported normal levels (Parati et al., 1980; Murialdo et al., 1989; Klimek et al., 1984). The LH response to LHRH has been reported as normal (Facchinetti et al., 1988) and reduced (Klimek et al., 1984; Murialdo et al., 1989), whilst an increased FSH response in both episodic and chronic cluster headache has been observed (Murialdo et al., 1989). A reduction in LH peaks over 24 hours has been demonstrated in both chronic and episodic cluster headache (Micieli et al., 1987), further indicating an abnormality in the central control of LH secretion.

Testosterone levels have been reported as normal (Nelson, 1978) and low (Kudrow, 1976; Klimek, 1982; Micieli et al., 1987) during the cluster period, the reported low levels possibly being secondary to reduced central LH production. There is evidence of increased responsiveness to exogenous testosterone in cluster headache sufferers compared to controls (Nicolodi et al., 1993) suggesting increased sensitivity to testosterone. Refractory cluster headache has been resolved by gonadotropin releasing hormone (GnRH) analogues, suggesting that suppression of the hypothalamo-pituitary-gonadal axis can restore a patient to the pain-free state (Nicolodi et al., 1993b).

### *Growth Hormone Axis*

There have been reports of abnormalities in the growth hormone axis in cluster headache. An abnormal evening growth hormone peak has been reported in episodic cluster headache (Chazot et al., 1984) and Klimek (1985) reported an increased growth hormone response to dopamine agonists, with a simultaneous reduction in prolactin. Because hypothalamic dopaminergic neurones have been shown to facilitate

growth hormone release and inhibit prolactin (Anden et al., 1967), these observations may point to increased hypothalamic dopaminergic tone in cluster headache (Leone and Bussone, 1993).

#### *Thyroid Axis*

Although basal thyroid stimulating hormone (TSH) levels are predominantly normal in cluster headache (Waldenlind and Gustafsson, 1987; Bussone et al., 1988; Leone et al., 1990) there is evidence of reduced TSH responsiveness to thyrotropin releasing hormone (TRH) in both episodic (Bussone et al., 1988; Leone et al., 1990) and chronic cluster headache (Waldenlind and Gustafsson, 1987) compared with healthy controls.

#### *Dopamine-Prolactin Axis*

The diurnal rhythm of prolactin has been reported as normal (Ferrari et al., 1979; Chazot et al., 1984) with a loss in prolactin release rhythms (Polleri et al., 1982; Ferrari et al., 1983). Waldenlind et al. (1987) demonstrated reduced 24-hour prolactin production in episodic cluster headache both in remission and during the cluster period, which was independent of sleep deprivation. This reduction in prolactin secretion may be related to increased dopaminergic tone in cluster headache (Waldenlind and Gustafsson, 1987). A blunted prolactin response to dopamine antagonists has been demonstrated in episodic cluster headache during a bout, (Klimek, 1985) and Bussone et al. (1988) demonstrated no alteration in prolactin response to TRH, which may indicate that dopaminergic pathways are specifically altered in cluster headache.

In summary, the literature suggests that there are alterations in all five hypothalamo-pituitary axes in patients with cluster headache, both in the resting state and during a bout, compared with controls as summarised in Table 1. It is conceivable that by studying the headache phenotypes characterised by specific perturbations in the hypothalamo-pituitary axis in functional pituitary tumours, further light may be shed on the precise biochemical mechanisms involved in primary headache. Further exciting and more direct evidence of the importance of hypothalamic activation in CH arises from functional imaging studies, which is now discussed.

**Table 1. Biochemical abnormalities in cluster headache**

<b>Axis</b>	<b>Basal Test</b>	<b>Dynamic Test</b>
<b>HPA</b>	<ul style="list-style-type: none"> <li>• Loss of diurnal variation</li> <li>• ↑ 24 h urinary cortisol</li> </ul>	<ul style="list-style-type: none"> <li>• ↓ response to stress</li> <li>• ↓ response to CRH</li> <li>• ↓ response to hypoglycaemia</li> <li>• N dexamethasone suppression</li> </ul>
<b>Gonadal</b>	<ul style="list-style-type: none"> <li>• N, ↓ LH</li> <li>• ↓ 24 h LH secretion</li> <li>• N, ↑ FSH</li> <li>• N, ↓ Testosterone</li> </ul>	<ul style="list-style-type: none"> <li>• N, ↓ LH response to LHRH</li> <li>• ↑ FSH response to LHRH</li> </ul>
<b>GH</b>	<ul style="list-style-type: none"> <li>• ↑ evening peak GH</li> </ul>	<ul style="list-style-type: none"> <li>• ↑ response to dopamine agonists</li> </ul>
<b>TSH</b>	<ul style="list-style-type: none"> <li>• N</li> </ul>	<ul style="list-style-type: none"> <li>• ↓ response to TRH</li> </ul>
<b>Prolactin</b>	<ul style="list-style-type: none"> <li>• N, ↓ 24 h secretion</li> </ul>	<ul style="list-style-type: none"> <li>• ↓ response to dopamine agonists</li> <li>• N response to TRH</li> </ul>

HPA = Hypothalamo-pituitary adrenal      N = normal

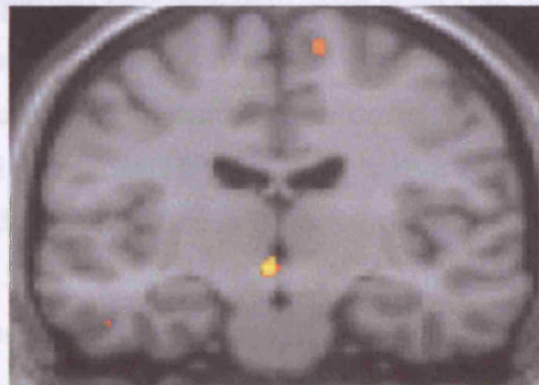
### *1.5.1.3 Functional Imaging Abnormalities*

Activation of the ipsilateral posterior hypothalamic grey matter has been demonstrated using PET scans during nitroglycerin-induced cluster headache attacks (May et al., 1998a). Evidence that this hypothalamic activation is specific to cluster headache and not a secondary marker of pain, is illustrated by the lack of this pattern of activation in migraine (Weiller et al., 1995) or experimentally induced ophthalmic division pain (May et al., 1998b). Voxel-based morphometric analysis of the structural T1-weighted MRI scans of 25 patients with cluster headache (May et al., 1999a) showed that cluster patients had an increase in hypothalamic volume compared to healthy controls, and the inferior posterior hypothalamus was the specific area of enlargement. This area closely correlates with the PET appearances and adds consistency to the view that this nucleus is involved in cluster headache (Figure 1).

Leone et al. (2001) reported a patient with intractable cluster headache who experienced a dramatic resolution in symptoms after stereotactic stimulation of the posterior hypothalamus. Subsequent to this, five patients have responded well to the insertion of deep brain hypothalamic stimulators (Franzini et al., 2003).

**Figure 1. Ipsilateral hypothalamic activation in cluster headache on PET**

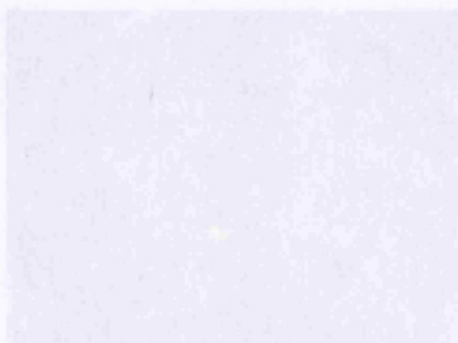
Taken from May et al. (1998a)



In summary, primary cluster headache appears to involve structural / functional disturbances of the hypothalamo-pituitary axis. As will be discussed in section 1.8.3, there are reported cases of cluster headache occurring in association with pituitary tumours (Milos et al., 1996; Tfelt-Hansen et al., 1982; Greve and Mai, 1988; Porta-Etessam et al., 2001), and a key question is whether this is due to functional or structural abnormalities within the pituitary gland.

**Figure 2. Ipsilateral hypothalamic activation in SUNCT on PET**

Taken from May et al. (1998)

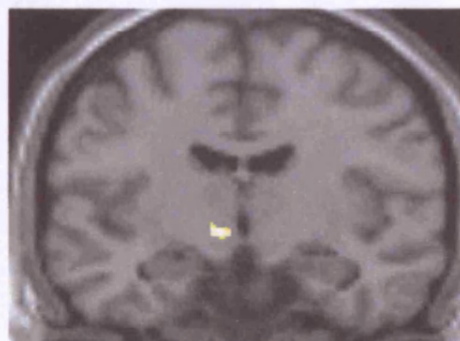


### **1.5.2 Short lasting Unilateral Neuralgiform headache attacks with Conjunctival injection and Tearing (SUNCT)**

The syndrome of short lasting unilateral neuralgiform headache attacks, conjunctival injection and tearing (SUNCT) is characterised by short attacks of pain, lasting between 5-250 seconds, with a mean duration of 49 seconds (Pareja et al., 1996). Patients are generally pain-free in between episodes (Pareja et al., 1996). The frequency of attacks varies from one per day to 30 attacks per hour (Pareja and Sjaastad, 1997), and episodes predominantly occur during the day unlike cluster headache where nocturnal attacks are common. Attacks can be precipitated by touching the face or scalp, eating, washing, shaving or talking, making it clinically very similar to trigeminal neuralgia (Pareja and Sjaastad, 1997). Functional imaging studies have suggested that SUNCT attacks involve the ipsilateral hypothalamus (May et al. 1999; Figure 2). SUNCT is clinically similar to trigeminal neuralgia, although the presence of a refractory period, the relative absence of cranial autonomic features, the shorter lasting attacks and response to carbamazepine are features that favour a diagnosis of trigeminal neuralgia rather than SUNCT (Matharu and Goadsby, 2002). SUNCT may also be confused with primary stabbing headache (Pareja et al., 1999). This refers to sharp jabbing pains, usually in the ophthalmic distribution of the trigeminal nerve, that are often associated with a concurrent primary headache syndrome. It is thought that primary stabbing headache represents a general reflection of abnormal trigeminal activity and does not in itself have a large pathophysiological significance. Primary stabbing headache is seen more commonly in women than men, and often responds to indomethacin (Pareja et al., 1999). As will be discussed in section 1.8.2, SUNCT has been described in association with pituitary tumours (Massiou et al., 2002; Levy et al., 2003a; Matharu et al., 2003).

**Figure 2. Ipsilateral hypothalamic activation in SUNCT on fMRI**

Taken from May et al. (1999)



### 1.5.3 Paroxysmal Hemicrania

Paroxysmal hemicrania lies between cluster headache and SUNCT in terms of the duration and frequency of headache attacks. Paroxysmal hemicrania is characterised by excruciating unilateral pain and autonomic features that last 10-30 minutes, the length of attack ranging from 2-45 minutes (Antonaci and Sjaastad, 1989). The frequency of attacks is typically greater than five per day, the reported range being 1-40 per day (Antonaci and Sjaastad, 1989). The female preponderance and dramatic response to indomethacin distinguish paroxysmal hemicrania from cluster headache. The differences between the short-lasting headaches are summarised in Table 2.

**Table 2. Clinical features of short lasting headaches.**

After Matharu et al. (2002)

<b>Trigeminal Autonomic Cephalgia (TAC)</b>				<b>Other</b>	
<b>Feature</b>	<b>Cluster Headache</b>	<b>Paroxysmal Hemicrania</b>	<b>SUNCT</b>	<b>Primary Stabbing Headache</b>	<b>Trigeminal Neuralgia</b>
<b>Pain</b>					
Type	boring	boring	stabbing	stabbing	stabbing
Severity	v. severe	v. severe	severe	severe	v.severe
Location	orbital	orbital	orbital	any	V2 / V3 > V1
Duration	15-180 mins	2-45 mins	15-120s	<30s	<1s
Frequency	1-8/day	1-40/day	1/day-30/hour	any	any
Autonomic	+	+	+	-	-

## 1.6 Chronic Daily Headache

Chronic daily headache is not a diagnostic term, but is useful in clinical practice in a similar way to the phrase 'anaemia', in that it describes a clinical phenomenon without having pathophysiological implications (Goadsby and Boes, 2002). Chronic daily headache is defined as headache on 15 days or more per month for more than three months (Goadsby and Boes, 2002). The first step in classifying chronic daily headache is to determine if it is a primary or secondary headache. Secondary headache implies an underlying pathology and the causes are numerous, including traumatic, inflammatory, infective or neoplastic (Goadsby and Boes, 2002; Table 3).

**Table 3. Classification of chronic daily headache.**

Adapted from Welch and Goadsby (2002)

<b>Primary</b>		<b>Secondary</b>
<b>≥4 h per day</b>	<b>&lt;4 h per day</b>	
Chronic migraine	Chronic cluster headache	<i>Post-traumatic</i> Head injury Iatrogenic Post-infectious Neoplastic
Chronic tension-type headache	Chronic paroxysmal hemicrania	<i>Inflammatory</i> Giant cell arteritis Sarcoidosis Behcet's
Hemicrania continua	SUNCT	<i>CNS infection</i>
New daily persistent Headache	Hypnic headache*	<i>Substance overuse</i>

\* Included for completion. A rare primary headache syndrome that awakes patient from sleep lasting 5 – 60 minutes with no autonomic features

### 1.6.1 Primary chronic daily headache



### *1.6.1.1 Chronic migraine*

Migraine may be viewed as an increased sensitivity to both internal and external stimuli during an exacerbation (Goadsby and Boes, 2002). In the past, chronic migraine, transformed migraine and chronic daily headache were used interchangeably and it has been suggested that the term chronic migraine is now used for patients with frequent headache on a migrainous basis (Welch and Goadsby, 2002). The term 'transformed migraine' was previously used to describe the observation of episodic migraine 'transforming' into a less discrete entity characterised by frequent headache with less obvious migrainous characteristics. This term does not acknowledge the difference between cause and association (Mathew et al., 1982). Welch et al. (2001) reported increased tissue iron levels in the periaqueductal gray matter (PAG) in patients with chronic migraine and analgesia overuse, suggesting a specific pathophysiological basis to this condition.

### *1.6.1.2 Chronic Tension-Type Headache*

Chronic tension-type headache is characterised by a featureless frequent headache. Tenderness of the pericranial muscles is the most commonly reported symptom in tension-type headache, possibly mediated by increased sensitivity of mechano-sensitive afferent neurones projecting to the dorsal horn of the trigeminal nucleus (Jensen and Olesen, 1996). Episodic tension-type headache may progress to chronic tension-type headache as a result of persistent and prolonged sensory input (Bendtsen and Ashina, 2000). Unlike migraine chronic tension-type headache is not associated with supraspinal dysmodulation of trigeminal pain transmission (Lipchik et al., 2000). CGRP levels are elevated in tension-type headache with throbbing (Ashina et al., 2000) and it may be more helpful to consider the latter as a form of migraine, particularly as there may be a therapeutic response to triptans (Lipton et al., 2000; Welch and Goadsby, 2002).

What is the point of this discussion in relation to pituitary tumour-associated headache and this thesis? The primary purpose is to recognise that patients with chronic daily headache in association with pituitary tumours may have concurrent chronic tension type headache or chronic migraine, and in order to parse out the specific role of the pituitary gland in patients presenting with headache, it is important to have a balanced perspective on the commoner causes of chronic daily headache.

### *1.6.1.3 Hemicrania continua*

Hemicrania continua is a rare primary headache characterised by unilateral side-locked pain, whose severity may wax and wane (Pareja et al., 2001). Hemicrania continua was a thought to be a featureless headache, but it is now recognised that migrainous features may be associated with this condition (Peres et al., 2002).

Hemicrania continua has a striking indomethacin-responsiveness in the absence of improvement with other non-steroidal anti-inflammatory agents. There is debate as to whether hemicrania continua can be safely diagnosed in a non-indomethacin responsive patient even if the other features are highly suggestive of hemicrania continua (Goadsby and Lipton, 2002). Case 2 in the Appendix (Levy et al., 2003a) describes a prolactinoma presenting with hemicrania continua after the administration of dopamine agonists, suggesting that the hypothalamo-pituitary axis may play a role in this headache phenotype.

In summary, there is a range of headache syndromes that appear to involve the hypothalamo-pituitary axis in their pathophysiology. The following part of the introduction summarises the headache syndromes that have been observed in association with pituitary tumours, and the potential mechanisms involved will be discussed.

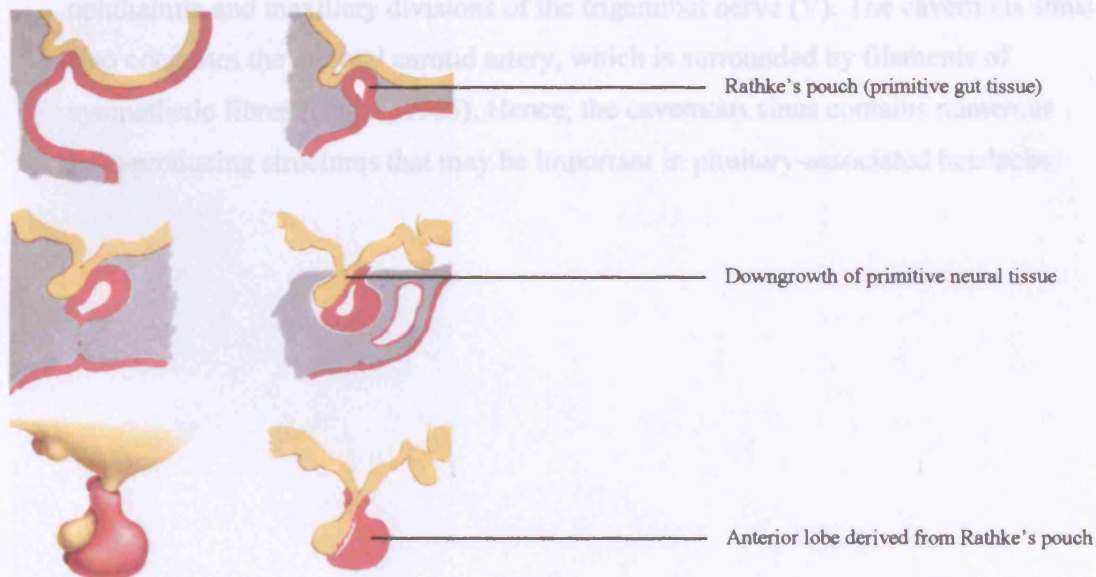
## 1.7 An Introduction to Pituitary Tumours

### 1.7.1 The Normal Pituitary Gland

The pituitary gland is a small structure, which sits in the pituitary fossa at the base of the brain. Embryologically, the pituitary gland is a combination of primitive gut and neural tissue (Dorton, 2000). The anterior pituitary gland is derived from an up-growth of gastrointestinal ectodermal tissue (Rathke's Pouch), which is pinched off to form the anterior lobe. The posterior lobe is derived from neural ectoderm, which extends from the floor of the primitive forebrain to lie adjacent to the anterior lobe. The anterior lobe gives off two ventral processes that surround the upper end of the neural stalk forming the 'pars tuberalis'. The mature pituitary is thus formed, consisting of the anterior lobe, which is primarily endocrinologically active glandular tissue, and the posterior lobe, which is primarily neural tissue (Figure 3).

**Figure 3. Development of the pituitary gland**

Taken from Netter (1965)



The pituitary stalk consists of neurones originating from the hypothalamus and the vascular hypophysial portal system, and this connection between the hypothalamus and pituitary gland comprises the hypothalamo-pituitary axis. The central dogma of endocrinology is that the hypothalamus communicates with the posterior pituitary via

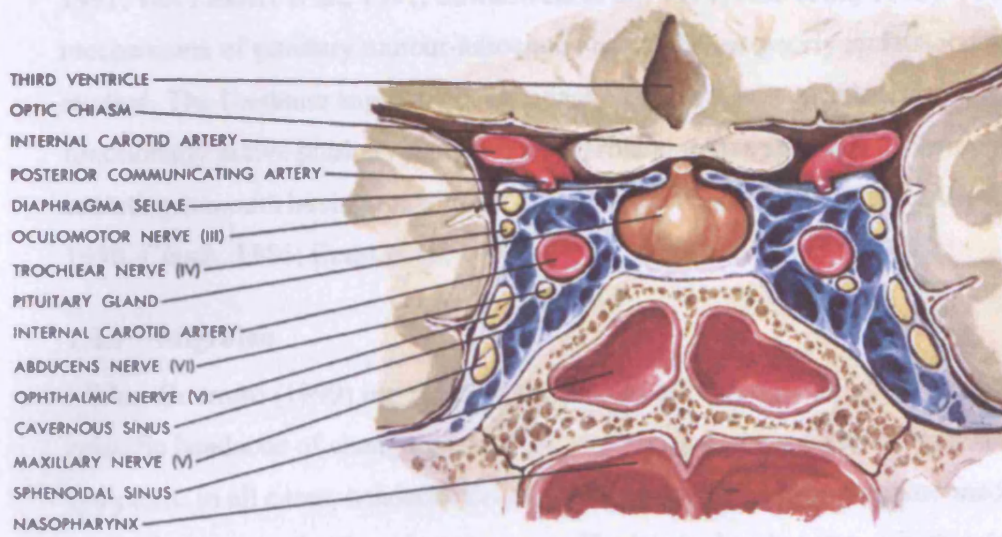
neural pathways, and with the anterior pituitary via the secretion of 'releasing hormones' into the portal system (Faglia and Ambrosi, 1992). Although the anterior pituitary is primarily composed of glandular tissue, there are data to show that each glandular cell is innervated by hypothalamic neurones containing substance P and CGRP, suggesting that the anterior lobe is under direct neural as well as humoral regulation (Ju and Liu, 1989; Ju and Zhang, 1990; Ju et al., 1993; Liu, 1995; Liu and Gao, 1998), which may be relevant to pituitary tumour-associated headache.

### **1.7.3 Anatomical Relations**

The adult pituitary gland measures approximately 12mm transversely, 8mm in the antero-posterior diameter, and 6mm in its vertical dimension (Figure 4). The roof of the pituitary fossa is formed by a circular fold of dura mater (the diaphragma sellae). The latter is pierced by a small central aperture, through which the pituitary stalk passes, and this separates the anterior part of the upper surface of the gland from the optic chiasm. Laterally, the pituitary gland is bounded on each side by the cavernous sinus and the structures within it. The lateral wall of the cavernous sinus contains the ophthalmic and maxillary divisions of the trigeminal nerve (V). The cavernous sinus also contains the internal carotid artery, which is surrounded by filaments of sympathetic fibres (Netter, 1965). Hence, the cavernous sinus contains numerous pain-producing structures that may be important in pituitary-associated headache.

**Figure 4. Anatomical relations of the pituitary gland**

Taken from Netter (1965)



## **1.8 Pituitary Tumours and Headache: a Clinical Review**

The prevalence of headache in pituitary tumours ranges from 33-72% (Comtois et al., 1991; van Lindert et al., 1991; Suwanwela et al., 1994; Abe et al., 1998). The mechanisms of pituitary tumour-associated headache are poorly understood and little studied. The literature suggests that headache is particularly problematic in functionally active pituitary tumours, with prolactinomas and growth hormone-secreting tumours having the highest reported prevalence of headache (Abe et al., 1998, Couch, 1986; Ezzat et al., 1994).

### **1.8.1 Migraine**

Millan-Guerrero (1999) reports 20 patients with micro-prolactinomas presenting with episodic headache of changing laterality, ten having had headache as their sole symptom. In all cases, headache disappeared upon receiving appropriate medical or surgical treatment for the microadenoma. The headache phenotypes in these cases are described as migrainous and the author comments that ‘this “migraine-type” pain is probably related to hormonal activity as it is difficult to relate to nerve terminal stimulation of the trigeminal nerve or stimulation of pain structures within the cavernous sinus...due to the very small dimensions of the tumour’. This sentiment is echoed by Gabrielli (2002) who reports the case of migraine without aura in a patient with a micro-prolactinoma. Having had eight attacks per month for 6 years, this patient’s headache disappeared within 2 months of bromocriptine therapy. The author argues, ‘as bromocriptine did not affect the size of the pituitary adenoma, a decrease in mass effect or traction on adjacent structures appears unlikely’. Lee (1990) reports a 47 year-old female with a five-year history of episodic migraine with aura, each episode lasting 2-3 hours and occurring once a month. After removal of the tumour, which was a non-functioning adenoma, the episodes of migraine with aura completely disappeared.

Shah and Frejj (1999) report a pituitary macroadenoma presenting with bilateral throbbing headache associated with photophobia that was dramatically relieved by sumatriptan, despite being unresponsive to analgesics including opioids. Pascual (2000) suggests that the analgesic effect of sumatriptan in this case could be due to ‘inhibition of the secretion of algescic peptides by the tumour’ such as calcitonin gene

related peptide (CGRP).

### **1.8.2 SUNCT**

Massiou et al. (2002) report SUNCT syndrome in two patients with prolactinomas presenting with bromocriptine-induced attacks. The second case had a micro-prolactinoma associated with attacks of pain after bromocriptine and lisuride administration. This is very similar to Case 1 described in this Appendix (Levy et al., 2003a). Ferrari et al. (1988) report a microprolactinoma presenting with ‘trigeminal neuralgia’ with pronounced autonomic features in response to bromocriptine therapy. However, this patient almost certainly had SUNCT syndrome, and the headache diagnosis is probably a reflection of the time of writing of the article when SUNCT syndrome was not yet described. Ferrari et al. (1988) acknowledge the association between the D<sub>2</sub> agonist and the headache onset, arguing that chemical or micro-vascular changes within the pituitary gland may have been responsible. Case 5 (Appendix) describes SUNCT in association with a macroprolactinoma (Matharu et al., 2003), which resolved soon after the administration of cabergoline, indicating that D<sub>2</sub> agonists may exacerbate or alleviate pituitary-associated SUNCT.

### **1.8.3 Cluster Headache**

Milos et al. (1996) describe a 37 year-old man presenting with left-sided cluster headache that was refractory to medical therapy. Five years after presentation, a diagnosis of acromegaly was made, and a left-sided macro-adenoma was confirmed. After transphenoidal removal of the tumour, ‘the patient (has) been absolutely free from further headache attacks’. Cluster headache has also been reported in the presence of prolactinoma (Tfelt-Hansen et al., 1982; Greve and Mai, 1988; Porta-Etessam et al., 2001; Menguzzi et al., 2003) and in each case, attacks resolved after treatment of the pituitary pathology. Although Greve & Mai describe their case as cluster headache, the high frequency of attacks (10 per day) and short duration (15 to 30 minutes) suggests that paroxysmal hemicrania may have been the diagnosis.

### **1.8.4 ‘Trigeminal Neuralgia’**

The classification of headache has evolved in recent years. As has been described in Ferrari et al.’s SUNCT case (1988), the term ‘trigeminal neuralgia’ may have been

incorrectly used for short-lasting stabbing headaches associated with pronounced autonomic features. Other authors have similarly reported 'trigeminal neuralgia' in association with pituitary adenomas (Friedman et al., 1982; Gelabert Gonzalez et al., 1990; Gazioglu et al., 2000), the symptoms resolving after appropriate management of the pituitary pathology. Friedman et al. (1982) argue that their case 'most likely had facial pain because of tumour invasion of the cavernous sinus', although they do not explain the episodic nature of the pain in this patient. On closer analysis of this case, attacks lasted 30 minutes to 3 hours and were associated with ipsilateral 'tearing..and increased nasal vasomotor activity', suggesting that a trigeminal autonomic cephalgia may have been present. Trigeminal neuralgia is characterised by very short lasting jabs of pain, typically in the maxillary or mandibular divisions of the trigeminal nerve, typically precipitated by minor irritation of areas innervated by the trigeminal nerve, such as touching the face or scalp, washing, chewing or talking and the accuracy of this diagnosis in the above cases should be taken with a degree of caution. A summary of the headache phenotypes associated with pituitary tumours is given in Table 4.



**Table 4. Headache phenotypes described with pituitary tumours**

Headache Phenotype	Author (date)	Tumour Type	Response to treatment (+) improved (-) exacerbated
<b>Migraine</b>	Gabrielli et al. (2002)	Prolactinoma	dopamine agonist (+)
	Shah & Frejj (1999)	NFA	sumatriptan (+)
	Millan Guerrero (1999)	Prolactinoma	dopamine agonist (+)
	Lee (1990)	NFA	hypophysectomy (+)
<b>SUNCT</b>	Levy et al. (2003)	Prolactinoma	dopamine agonist (-)
	Matharu et al. (2003)	Prolactinoma	dopamine agonist (+)
	Massiou et al. (2002)	Prolactinoma	dopamine agonist (-)
<b>Cluster Headache</b>	Porta-Etessam (2001)	Prolactinoma	dopamine agonist (+)
	Milos et al. (1996)	GH	hypophysectomy (+)
	Greve & Mai** (1988)	Prolactinoma	dopamine agonist (+)
	Tfelt-Hansen et al. (1982)	NFA/ Prolactinoma††	hypophysectomy (+)
<b>'Trigeminal Neuralgia'</b>	Gazioglu et al. (2000)	Mixed GH and prolactin secreting	hypophysectomy (+)
	Ferrari et al. (1998)*	Prolactinoma	dopamine agonist (-)
	Galabert Gonzalez et al. (1990)	Craniopharyngioma	hypophysectomy (+)
	Friedman et al.	NFA	hypophysectomy and radiotherapy (+)

\* SUNCT was the probable diagnosis in this case

\*\* Paroxysmal hemicrania probable diagnosis

### 1.8.5 Octreotide

Musolino et al. (1990) report two cases of acromegalic headache displaying dramatic improvement after the administration of octreotide. Case 2 of Musolino's paper presents with a "pinprick" sensation accompanied by lacrimation, redness of the eye, and nasal congestion as seen in cluster headache'. In these cases, there was an immediate analgesic response to octreotide, although with time, the length of analgesia 'decreased progressively to the point that administration of the drug was necessary every 2 hours, with recrudescence of the pain after this interval'. This description of octreotide tolerance and dependency is similar to our own Case 5 (Appendix; Levy and Goadsby, 2004) and other observations (Popovic et al., 1988; May et al., 1994). Pascual et al. (1991) report a patient with acromegaly who had intractable headache 'unrelated to tumour size', which dramatically resolved with

octreotide. They suggest that the analgesic action of octreotide ‘could not be related to a simultaneous decrease in growth hormone or prolactin...and furthermore, octreotide analgesic effects cannot be explained by tumour shrinkage’. They also comment that ‘by understanding how octreotide relieves pain in these cases, the pathophysiology of pituitary adenoma-related headache could be clarified.’ In this excellent paper, they provide evidence that the analgesia is not due to the placebo effect, by double-blind placebo controlled administration of octreotide, and they also show a lack of reversibility with naloxone, suggesting that octreotide-induced analgesia is not via opioidergic mechanisms.

Williams et al. (1987) present six patients with pituitary tumour-associated headache, five of whom had acromegaly and one a prolactinoma. All patients were withdrawn from analgesics and admitted to hospital. Using a visual analogue scale, patients scored their headache after double blind administration of octreotide 100 µg or placebo. Three of the five acromegalics, and the single prolactinoma case, responded dramatically to octreotide but not placebo. The authors comment that ‘though headache is traditionally attributed to the space occupying effects of the tumour, other factors must contribute as severity of headache correlates poorly with size...(and) analgesia with octreotide occurs too rapidly to be explicable by tumour shrinkage..and...an interesting possibility is that octreotide may suppress secretion by the pituitary tumour of “algesic” peptides which may cause pain’.

Schmidt et al. (1993) report a double-blind placebo controlled analgesic response to octreotide 100 µg in two acromegalics. They observed pain relief within 4 – 15 minutes that lasted 2 – 8.5 hours after injection with no evidence of tachyphylaxis.

Webb et al. (1989) report an acromegalic female whose disease ‘remained active and induced invalidating headache in spite of previous treatment with surgery and radiotherapy’. After the administration of octreotide, ‘headache was improved in a matter of minutes, even if normalisation of hormone hypersecretion was not demonstrated’. This uncoupling of growth hormone and headache in response to octreotide has been described earlier in our own Case 3 (Appendix; Levy et al., 2003b) suggesting that octreotide has a specific analgesic effect unrelated to growth

hormone suppression. Webb et al. (1989) acknowledge that octreotide may be a ‘therapeutic option in patients with headache unresponsive to common analgesics’. A discussion of the potential mechanisms of octreotide-induced analgesia occurs in section 1.18. A summary of the reports of octreotide-analgesia and octreotide dependency is given in Table 5.

**Table 5. Octreotide analgesia and dependency in pituitary tumours**

<b>Author (date)</b>	<b>Placebo-controlled analgesic response (+ / -)</b>	<b>Octreotide dependency (+ / -)</b>
Levy et al. (2003)†	-	-
May et al. (1994)	-	+
Schmidt et al. (1993)	+	-
Pascual et al. (1991)†	+	-
Musolino et al. (1990)	-	+
Webb et al. (1989)†	-	-
Popovic (1988)	-	+
Williams et al. (1987)	+	-

† In these cases, analgesic action of octreotide was not coupled to growth hormone suppression

## **1.9 Structural Explanations for Headache in Pituitary Tumours**

### **1.9.1 Cavernous Sinus Invasion**

The cavernous sinus contains pain-producing structures and it is not unreasonable to suppose that distortion of these would cause headache. The walls of the cavernous consist of dura mater which are densely innervated by trigeminal afferent neurones (Levy and Strassman, 2002), and irritation of the latter could cause trigeminal irritation. The walls of the venous plexi within the cavernous sinus are homologous to the cerebral venous sinuses, and activation of nociceptive pathways with stimulation of the superior sagittal sinus is well documented experimentally (Goadsby et al., 1991). The internal carotid artery is densely innervated with autonomic fibres, and encasement of the carotid artery by an invasive pituitary tumour could explain pain (Friedman et al., 1982). Stimulation of cranial arterial structures, especially the middle meningeal artery, is known to activate the trigeminal system (Hoskin et al., 1999). The ophthalmic and maxillary branches of the trigeminal nerve lie within the cavernous sinus, and direct irritation of these structures would be expected to cause activation of the trigeminovascular system.

#### *Non-pituitary para-sellar lesions*

Evidence that pituitary tumour-associated headache may simply be due to structural irritation of the cavernous sinus comes from reported cases of non-pituitary parasellar lesions invading the cavernous sinus.

Tolosa-Hunt syndrome is a rare disorder caused by granulomatous infiltration into the cavernous sinus, presenting with severe headache and cranial nerve palsies (Tolosa 1954; Hunt et al. 1961). Hannerz et al. (1984) reported that conventional imaging techniques may not show an abnormality in the cavernous sinus, although venography may show occlusion or slowing of venous drainage within the cavernous sinus and ophthalmic veins. Tolosa-Hunt can present with severe headache syndromes with minimal changes on MRI and has led some groups to believe that the *prima facie* pathology in cluster headache lies within the cavernous sinus. To support this, there is evidence of abnormal orbital phlebography in patients with cluster headache (Hannerz

et al., 1987), SUNCT syndrome (Hannerz et al., 1992; Kruszewski, 1992) and chronic paroxysmal hemicrania (Antonaci, 1994). Magnetic Resonance Angiography (MRA) performed during spontaneous attacks of cluster headache has shown marked dilation of the ophthalmic artery ipsilateral to the pain (Waldenlind et al., 1993; Ekbom and Greitz, 1970). Harbedo et al. (1991) argue that the abnormal orbital phlebography seen in cluster headache, the fact that potent vasodilators such as nitroglycerine can trigger an attack, and the finding that cluster headache patients have narrow middle cranial fossae, and by extrapolation slower drainage of the cavernous sinuses (Afra et al., 1998), are evidence that cluster headache is a disease of the cavernous sinus. PET studies during induced cluster attacks have shown pooling of tracer bilaterally within the cavernous sinus, with increased pooling ipsilateral to the pain (May et al., 1999a) which is thought to represent increased flow in the cavernous section of the internal carotid arteries.

Those that favour a central rather than peripheral cause for cluster headache argue that MRI studies of cluster patients show no pathological changes in the cavernous sinuses (Sjaastad and Rinck, 1990). Also, PET studies show activation of the cavernous sinus in capsaicin-induced experimental head pain, which is strong evidence that cavernous sinus changes are secondary to activation of the trigeminovascular system (May et al., 1998b).

Other non-pituitary para-sellar lesions involving the cavernous sinus have been associated with trigeminal autonomic cephalgias. Greve & Mai (1988) describe a cluster patient with a cavernous carotid artery aneurysm. Sjaastad et al. (1988) report a patient with cluster headache in a patient with an aneurysm of the cavernous section of the anterior communicating carotid artery. Hannerz (1989) reports a parasellar meningioma, and Narbone et al. (1991) present a calcified third ventricle lesion of unknown pathology, both patients presenting with cluster headache. Vijayan (1992) reports a 34 year-old female with chronic paroxysmal hemicrania who had a tumour in the sella turcica that invaded the cavernous sinus.

Hence the cavernous sinus plays a prominent part in both primary and secondary cluster headache, and many physicians have not looked beyond this structure when considering the mechanism of headache in pituitary tumours. However, the only

systematic study that has looked at the relationship between cavernous sinus invasion and headache in pituitary tumour found there to be no association (Abe et al., 1998). Moreover, there is no adequate accepted explanation for the severe headache syndromes caused by micro-adenomas, nor an explanation for the immediate somatostatin-responsiveness of pituitary tumour-associated headache without reduction in tumour size. Whilst clinical experience suggests that there is a sub-group of patients who have headache as a significant problem with cavernous sinus invasion (de Groot & Jameson, 2000) there are patients who do not suffer with headache despite extensive invasion into this structure. Hence, there may be some other factor(s) involved, such as the properties of the tumour itself.

### **1.9.2 Miscellaneous Structural Explanations**

In addition to cavernous sinus invasion, traction and displacement of the diaphragma sella by suprasellar extension of a pituitary tumour is commonly used to explain headache (Wirth and Van Buren, 1971; Dalessio, 1978), although Abe et al.'s study (1998) showed no relationship between pituitary tumour size and headache. Arafah et al. (2000) suggest that intrasellar pressure is important in the aetiology of headache. This group looked at 49 patients presenting with pituitary tumours, of which 25 had headache. They measured intra-sellar pressure peri-operatively, using a pressure transducer, and found that an association between raised intra-sellar pressure and headache. They also suggest that the headache observed in micro-adenomas is due to pressure within a small volume, rather than volume itself. It is conceivable that, for example in dopamine agonist induced headache in prolactinomas (Ferrari et al., 1988; Massiou et al., 2002; Levy et al., 2003a), there is some critical change in pressure within the lesion that causes headache, rather than a biochemical explanation. Other authors explain the episodicity of pituitary tumour-associated headache as being due to a 'ball-valve' effect that could cause transient structural abnormalities after a critical change in volume or pressure that eventually resolves (Lee, 1990).

## **1.10 Biochemical Explanations for Headache in Pituitary Tumours**

Some groups believe that structural factors alone are not a sufficient explanation for pituitary tumour-associated headache, arguing that pituitary tumours may secrete a nociceptive peptide (Pascual, 2000; Williams et al., 1986). Several candidate peptides exist, which are now discussed.

### **1.11 Calcitonin Gene Related Peptide**

Calcitonin gene related peptide (CGRP) is a 37- amino acid peptide that is widely expressed in the central and peripheral nervous system. CGRP acts via G protein-coupled receptors (CGRP<sub>1</sub> and CGRP<sub>2</sub> receptors) and is thought to have diverse biological activities (van Rossum et al., 1997). CGRP-immunoreactive cells constitute 40-50% of dorsal root ganglia cells (Gibson et al., 1984a), and are the major site of termination of nociceptive neurones and are abundant brain areas believed to be important in nociceptive processing such as the hypothalamus, periaqueductal grey, nucleus raphe magnus and thalamus (van Rossum et al., 1997).

#### **1.11.1 CGRP and the pituitary**

The role of CGRP in the anterior pituitary is unclear, although there are numerous CGRP binding sites in the hypothalamus and anterior pituitary gland (Tschopp et al., 1985; Wimalawansa et al., 1987). CGRP-immunoreactive nerve fibres have been demonstrated by electron-microscopy in the anterior pituitary gland and direct neural synapses have been shown to be in contact with corticotrophs, somatotrophs and lactotrophs (Ju, 1999). The origin of these nerve fibres has not been unequivocally determined, although Ju (1997) reports that they may originate from trigeminal or hypothalamic sources.

#### *CGRP and growth hormone*

CGRP has been shown to have stimulatory and inhibitory effects on growth hormone production. Tannenbaum and Goltzman (1985) demonstrated that CGRP suppresses growth hormone release when injected intracerebroventricularly. This was confirmed by Netti et al (1989), who showed suppression of growth hormone release after intracarotid injection of CGRP. Fahim et al. (1990) demonstrated that intracerebroventricular injection of CGRP suppressed growth hormone release, whilst

the injection of highly specific antiserum caused a transient elevation in growth hormone. However, Nakamura et al. (1998) found that the addition of CGRP to cultured human and rodent somatotrophs caused an increase in growth hormone secretion both with non-tumorous and pituitary adenoma cells. These data suggest that CGRP acts on growth hormone in an inhibitory manner when applied to central nervous structures, whilst acting as a growth hormone secretagogue when added locally. Netti et al. (1989) suggest that CGRP exerts its central inhibitory action on growth hormone via stimulation of somatostatin release.

#### *CGRP and prolactin*

CGRP has both inhibitory and neutral effects on prolactin secretion. Prolactin levels are inhibited when CGRP is added to cultured and TRH-stimulated pituitary cells (Shah et al., 1988). Netti et al. (1989) found no alteration in prolactin after intracerebroventricular injection of CGRP, although Fahim et al. (1990) found inhibition of prolactin secretion. The prolactin response to stress is blunted in rodents after subcutaneous or intraperitoneal CGRP infusion (Elie et al., 1990).

#### *CGRP and gonadotropins*

Gon et al. (1990) demonstrated CGRP immunoreactivity in gonadotrophs in developing rat embryos and neonates, particularly increasing post-natally between days 5 and 14. Adult rat gonadotrophs were found to have low CGRP immunoreactivity, which was increased during lactation and by oestrogen administration. Van Leeuwen et al. (1992) studied CGRP levels in 8 women with premenstrual syndrome and found no significant fluctuations in CGRP throughout the menstrual cycle. Wang et al. (1994) found intravenous CGRP administration to cause an inhibition of serum LH and testosterone in rodents.

#### *CGRP and thyrotrophin*

Intravenous administration of CGRP with calcitonin reduces the ability of TSH to stimulate thyroxine production in mice (Ahren, 1989). Hanna et al. (1995), having observed that the administration of calcitonin causes an increase in  $\alpha$ -subunit TSH-secreting tumours in rats, characterised a high affinity binding site for CGRP in the rat  $\alpha$ -TSH thyrotroph cell line. The finding of a high affinity binding site for CGRP has



been demonstrated in mouse thyrotroph cell lines, further implicating a functional role for CGRP in thyrotroph activity (Perry et al., 1997).

#### *CGRP and adrenocorticotrophin*

Kovacs et al. (1995) reported increased plasma cortisol levels following intracerebral CGRP injection, the responses being blocked by pre-treatment with CRH antiserum and concluded that CGRP causes pituitary-adrenal activation via CRH (Kovacs et al., 1995). Iino et al. (1998) demonstrated a stimulatory effect of CGRP on ACTH in cultured rat pituitary cells. They observed both a direct effect on basal ACTH production, and an additive effect with CRH (Iino et al., 1998). Dhillon et al. (2003) injected CGRP intracerebroventricularly and found this to stimulate hypothalamic CRH production.

#### *CGRP and pituitary tumours*

CGRP causes increased growth hormone release when added to somatotroph adenomas (Nakamura et al., 1998). Wimalawansa et al. (1994) has shown an increased expression of CGRP in pituitary adenomas, with preferential expression in growth hormone- and prolactin- secreting lesions (personal communication).

A summary of the effects of CGRP on the hypothalamo-pituitary axis is shown in Table 6.

**Table 6. Effects of CGRP on the HP axis**

<b>HP axis</b>	<b>CGRP administration</b>	<b>Effect</b>	<b>Reference</b>
<b>GH</b>	<i>in vitro</i>	↑	Nakamura et al., 1998
	Central injection	↓	Tannenbaum et al., 1985; Netti et al., 1989; Fahim et al., 1990
<b>Prolactin</b>	<i>in vitro</i>	↓	Shah et al., 1988
	Central injection	↓→	Fahim et al., 1990, Netti et al., 1989
	s/c and iv infusion	↓	Elie et al., 1990
<b>FSH/LH</b>	iv infusion in male rat	↓	Wang et al., 1994
<b>TSH</b>	iv infusion with CGRP + calcitonin in mouse	↓	Ahren, 1989
<b>ACTH</b>	<i>in vitro</i>	↑	Iino et al., 1998
	Central injection	↑	Dhillon et al., 2003

### 1.11.2 CGRP and headache

CGRP-containing sensory nerve fibres innervate cerebral arteries in animals (Uddman et al., 1985) and humans (Edvinsson et al., 1994; Edvinsson et al., 1987).

Pharmacological experiments have shown a pronounced vasodilatory effect of CGRP on cerebral arteries (Jansen et al., 1986). CGRP-immunoreactive fibres are present within the walls of the cerebral venous sinuses, and 'free' within the tissue matrix of the dura mater (Keller and Marfurt, 1991). The majority of these fibres derive from sensory neurons of the trigeminal ganglion.

A rise in CGRP has been demonstrated in patients undergoing thermocoagulation for trigeminal neuralgia (Goadsby et al., 1988). External jugular venous samples have shown a significant increase in CGRP during the headache phase of migraine with and without aura (Goadsby et al., 1990). CGRP levels returned to normal after headache resolution caused by sumatriptan administration (Goadsby and Edvinsson, 1993). Gallai et al. (1995) demonstrated increased CGRP levels during attacks of migraine with and without aura, with a greater increase in patients with aura.

Goadsby et al. (1994) showed CGRP levels to rise significantly during a cluster headache attack, as was observed by Fanciullacci et al. (1995) during nitroglycerin-induced cluster attacks. In both studies, spontaneous or sumatriptan-induced remission of attacks was associated with normalisation of CGRP levels strongly implicating a role for CGRP in cluster headache (Goadsby and Edvinsson, 1994; Fanciullacci et al., 1995).

Ashina et al. (2000) demonstrated elevated CGRP levels in chronic tension type headache associated with throbbing. CGRP was not demonstrated in patients with non-pulsating tension-type headache.

There is currently much interest in CGRP antagonists as potential new treatments for migraine and other primary headache disorders. The preliminary evidence that they may be efficacious is encouraging (Ramadan, 2001; Moreno et al., 2002; Olesen et al., 2004).

## **1.12 Substance P**

Substance P (SP) is an 11-amino acid peptide that may play an important role in nociceptive transmission (Harrison and Geppetti, 2001). SP is part of the tachykinin family, along with neurokinin A and neurokinin B, which share a similar chemical structure (Otsuka and Yoshioka, 1993). SP acts on a G-protein coupled receptor (the NK<sub>1</sub> receptor) and can also act on the receptors specific for neurokinin A and neurokinin B, termed NK<sub>2</sub> and NK<sub>3</sub> receptors, respectively (Regoli et al., 1994). SP is widely distributed within the central and peripheral nervous system and is frequently co-localised with CGRP in nociceptive areas (Ribeiro-da-Silva and Hokfelt, 2000). It is present in high concentrations in the dorsal horn (particularly laminae I and II), dorsal ganglion and dorsal root of spinal neurones (Ribeiro-da-Silva and Hokfelt, 2000) and is abundant in other areas of the central nervous system important in pain modulation such as the hypothalamus, periaqueductal grey, nucleus raphe magnus and the thalamus (Ribeiro-da-Silva and Hokfelt, 2000). The co-localisation of CGRP and SP may either reflect a necessary interaction between these peptides in nociceptive transmission, for example CGRP has been shown to inhibit the breakdown of SP (Le Greves et al., 1985), or it may preserve its own unique biological effect (Snijdelaar et al., 2000). Capsaicin, the irritant chemical in red-hot chilli peppers, acts on the

Vanilloid 1 (VR1) receptor (Szallasi and Blumberg, 1999) and has been shown to have a direct and selective stimulatory effect on C- and A $\delta$ - nociceptive fibres via the release of SP (Holzer, 1991).

### **1.12.1 Substance P and the pituitary**

SP is present in the hypothalamo-pituitary axis (Jessop et al., 1992) and is found within the anterior pituitary gland with its receptors (Larsen et al., 1989).

#### *Substance P and growth hormone*

Kato et al. (1976) demonstrated that SP is stimulatory to growth hormone secretion in the rat, which was confirmed by Rivier et al. (1977). Central administration of SP elevated growth hormone in ovariectomised rats (Vijayan and McCann, 1980) although other groups have shown an inhibitory effect (Arisawa et al., 1989a). Houben and Deneff (1993) demonstrated that that low doses of *in vitro* SP caused inhibition of growth hormone, whilst higher doses cause significant stimulation, which may explain the above differences. Systemic infusion of SP in humans causes elevation of basal growth hormone, and increases the stimulatory effect of GHRH (Coiro et al., 1992). However, Houben and Deneff (1993) failed to demonstrate an alteration in growth hormone release after *in vitro* administration of SP antagonists. Immunohistochemical studies have shown that the majority of SP positive cells in the rat anterior pituitary also contain growth hormone (Brown et al., 1991) and there is electron microscopic demonstration that SP-containing fibres are in direct contact with somatotrophs in primates (Ju, 1999) supporting a functional interaction between SP and growth hormone.

#### *Substance P and prolactin*

Intravenous infusion of SP in rats was initially shown to elevate serum prolactin (Kato et al., 1976) a finding that has also been shown after the *in vitro* administration of SP to cultured rat pituitary cells (Vijayan and McCann, 1980). Central injection of SP into the third ventricle (Vijayan and McCann, 1979; Eckstein et al., 1980) and the medial pre-optic area (Picanco-Diniz et al., 1990) resulted in an elevation of serum prolactin. Similar to its effect on growth hormone, low doses of SP may have an inhibitory effect on prolactin, whilst higher doses may be stimulatory (Arisawa et al., 1990). Larsen et al. (1992) demonstrated that a SP analogue binds specifically to

lactotrophs *in vitro* (Larsen et al., 1992). Traczyk et al. (1992) demonstrated a stimulatory effect of SP on prolactin when injected intracerebroventricularly. The withdrawal of dopamine has been shown to prolong SP-induced translocation of protein kinase C isozymes and subsequent release of prolactin in rat pituitary lactotrophs (Mau, 1997). Duvilanski et al. (2000) demonstrated that a specific NK<sub>1</sub> receptor antagonist reduces the TRH-stimulation of prolactin, suggesting that SP plays an important role in TRH-mediated prolactin release.

#### *Substance P and gonadotropins*

Intracerebroventricular injection of SP was initially shown to stimulate LH release in rats (Vijayan and McCann, 1979) whilst SP antagonists reduced serum LH and FSH (Arisawa et al., 1990b). Intravenous infusion of SP does not affect serum LH or FSH (Arisawa et al., 1990b) suggesting that SP acts centrally to release LHRH. There is a direct synaptic contact between SP fibres and cell bodies containing LHRH in the rat hypothalamus (Hoffman, 1985; Tsuruo et al., 1991). Serum levels of SP are known to fluctuate with the menstrual cycle in rats (Jakubowska-Naziemblo et al., 1985), and levels of SP within the anterior gland increase following ovariectomy (Coslovsky et al., 1984) further suggesting a functional link with gonadal hormones. High affinity SP receptors have been identified in the human anterior pituitary, and intravenous SP infusion stimulates LH release (Coiro et al., 1992). There is evidence in primates that endogenous SP potentiates the preovulatory LH and FSH surges during the menstrual cycle (Kerdelhue et al., 2000).

#### *Substance P and thyrotrophin*

Intracerebroventricular injection of SP causes an elevation in serum TSH in oestrogen-primed rats (Arisawa et al., 1989b). Brown et al. (1990) found no increase TSH secretion after *in vitro* administration of SP, although Moura et al. (1999) observed a significant increase in pituitary TSH release following treatment of incubated hemi-pituitaries with SP. There is an increase in rat pituitary SP following thyroidectomy, and a decrease following thyroxine administration (Aronin et al., 1984; Aronin et al., 1986; Jones et al., 1994). This observation is unaffected by transection of the pituitary stalk (Aronin et al., 1988). Intravenous infusion of incremental doses of SP did not influence TSH secretion in humans, suggesting that it may have no effect on TSH production outside the blood-brain barrier (Coiro et al.,

1995). SP-immunoreactive varicosities have been observed in close proximity to thyrotropes in primates (Ju and Liu, 1989; Liu and Gao, 1998) and there is co-secreted with TSH in both rats (Arita et al., 1994) and humans (Roth and Krause, 1990)

#### *Substance P and adrenocorticotrophin*

Chowdrey et al. (1990) demonstrated that intracerebroventricular SP injection inhibits ACTH release from the rat anterior pituitary. SP-immunoreactive neurones are present in the paraventricular nucleus of the hypothalamus, which is the main area for CRH production and release. Larsen et al. (1993) demonstrated that intracerebroventricular administration of a SP antagonist increases serum ACTH and cortisol and CRH mRNA in the paraventricular nucleus, suggesting that SP has a tonic inhibitory effect on hypothalamic CRH production. Melzig et al. (1995) demonstrated that the addition of substance P to pituitary tumour cells inhibits CRH-induced ACTH release, further indicating an inhibitory action of SP. Adrenalectomy can reduce pituitary SP content by 50% (Jones et al., 1990) and SP immuno-reactive fibres are in direct contact with anterior pituitary cells in the dog, about 60% of contacts occurring with corticotrophs (Ju and Zhang, 1990). Desiderio et al. (1993) observed human ACTH-secreting pituitary tumours to contain significantly higher levels of SP than non ACTH-secreting adenomas. A summary of the effects of SP on the hypothalamo-pituitary axis are shown in Table 7.

**Table 7. Effects of substance P on the HP axis**

<b>HP axis</b>	<b>Substance P administration</b>	<b>Effect</b>	<b>Reference</b>
<b>GH</b>	i.v infusion in rat	↑	Kato, 1976; Rivier, 1977
	Central injection (high dose)	↑	Vijayan, 1979; Houben, 1993
	Central injection (low dose)	↓	Arisawa, 1989; Houben, 1993
	iv infusion in humans	↑	Coiro, 1992
	<i>in vitro</i>	→	Houben, 1993
<b>prolactin</b>	iv infusion rats	↑	Kato, 1976
	<i>in vitro</i>	↑	Vijayan, 1980
	Central injection (high dose)	↑	Traczyk, 1992; Picanco-Diniz, 1990
	Central injection (low dose)	↓	Arisawa, 1990
<b>FSH/LH</b>	Central injection	↑	Vijayan, 1979
	iv infusion rats	→	Arisawa, 1990
<b>TSH</b>	Central injection	↑	Arisawa, 1989
	<i>in vitro</i>	→	Brown, 1990
<b>ACTH</b>	Central injection	↓	Chowdrey, 1990; Larsen, 1993

### 1.12.2 Substance P and headache

As with CGRP, SP-immunoreactive fibres richly innervate the cerebral arteries of animals (Uddman et al., 1985) and humans (Edvinsson et al., 1994) and this is supported by radioimmunoassay (Edvinsson et al., 1987) and pharmacological data (Jansen et al., 1986). The pattern of distribution of SP appears to be similar to CGRP in its innervation of the venous sinuses and the dura mater (Keller and Marfurt, 1991).

SP increases with CGRP in patients undergoing thermocoagulation for trigeminal neuralgia (Goadsby et al., 1988) but does not rise during migraine with and without aura (Goadsby et al., 1990; Gallai et al., 1995) or cluster headache attacks (Goadsby and Edvinsson, 1994; Fanciullacci et al., 1995). Unlike CGRP, SP levels were not found to change in patients with throbbing chronic tension type headache (Ashina et al., 2000).

The effect of neurokinin-1 (NK<sub>1</sub>) receptor antagonists has been investigated on animal models of trigeminal nociception and in clinical trials. Polley et al. (1997)

demonstrated that a potent NK<sub>1</sub> receptor antagonist (GR205171) resulted in a dose-dependent inhibition of plasma protein extravasation in dura mater, in response to electrical stimulation of the trigeminal ganglion. Shepherd et al. (1995) demonstrated that the non-peptide NK<sub>1</sub> receptor selective antagonist (CP-99,994) causes peripheral blockade of dural extravasation and central inhibition of nociceptive pathways, implying potential anti-migraine activity. However, Goadsby et al. (1998) found that SP blockade with GR205171 did not alter central trigeminal activity after superior sagittal sinus stimulation. Clinical trials investigating the potential use of NK<sub>1</sub> receptor antagonists in acute migraine have been disappointing. Goldstein et al. found no useful effect in the acute (1997) or preventive (2001) treatment of migraine. Diener et al. (2003) similarly found no effect of RPR100893, a SP-antagonist, in the acute treatment of migraine. Other negative studies investigating the intravenous use of NK<sub>1</sub> receptor antagonists in acute migraine include those from Connor et al. (1998) and Norman et al. (1998).

### **1.13 Neuropeptide Y**

Neuropeptide Y (NPY) is a 36-amino acid peptide and functions as a neurotransmitter and neuromodulator (Adrian et al., 1983). It mediates its actions via six NPY receptors (Y<sub>1</sub>-Y<sub>6</sub>), consisting of seven transmembrane G-protein coupled receptors. NPY is highly expressed in the hypothalamus, periaqueductal gray area, thalamus, trigeminal ganglion and dorsal horn of the spinal cord (Gibson et al., 1984b) suggesting a role in nociception. Both the Y and the Y<sub>1</sub> receptor are upregulated in rat spinal cord and dorsal root ganglia in response to pain (Ji et al., 1994). Intrathecal administration of NPY facilitates nociceptive neurones at low doses and inhibits them at higher doses (Xu et al., 1994). Similarly, intracerebroventricular administration is pro-nociceptive at low doses and antinociceptive at higher doses (Mellado et al., 1993). Other groups have shown only anti-nociceptive effects of NPY (Hua et al., 1991; Broqua et al., 1996).

#### **1.13.1 Neuropeptide Y and the pituitary**

NPY is present in abundance in the hypothalamo-pituitary axis (Ciofi et al., 1990; Byrne et al., 1992; Dumont et al., 1992) and an exhaustive discussion of its role is outside the scope of this thesis. It is thought to have an important role in energy homeostasis and is a potent stimulator of feeding in animals (Gehlert, 1999).



### *Neuropeptide Y and growth hormone*

McDonald et al. (1985) demonstrated that intracerebroventricular injection of NPY inhibits growth hormone secretion in ovariectomised rats, and stimulates it when applied to anterior pituitary cells *in vitro*. Adams et al. (1987) found NPY inhibited growth hormone secretion when added to cultured human pituitary somatotrophic tumours, implying a different action on tumorous somatotrophs compared to non-tumorous rat pituitary. In contrast, the intravenous infusion of NPY into humans with prolactinomas caused an elevation in serum growth hormone (Watanobe and Tamura, 1996). Watanobe and Tamura (1997) also demonstrated that intravenous NPY given to acromegalics had a stimulatory effect in patients with pure somatotroph tumours and an inhibitory effect in somato-mammotrophs, thus contradicting the *in vitro* findings. The inhibitory action of centrally-administered NPY on growth hormone secretion in rats may be mediated via Y<sub>1</sub> and Y<sub>2</sub> receptors, an effect that is abolished after deafferentation of the hypothalamus (Suzuki et al., 1996). Growth hormone secretagogues have been shown to inhibit growth hormone at supra-physiological doses, an effect that is probably mediated by NPY (Korbonits et al., 1999). Despite the inhibitory effect of central administration of NPY in the rat, it appears to have a stimulatory effect on growth hormone release in the cow (Thomas et al., 1999) and the ewe (Morrison et al., 2003).

### *Neuropeptide Y and prolactin*

Centrally administered NPY may stimulate prolactin release at low doses and inhibit its secretion at high doses (Chao et al., 1987). After the central injection of several NPY receptor agonists in the lateral ventricle of the rat, Garcia de Yebenes et al. (1995) demonstrated that Y<sub>2</sub> receptor agonists increased prolactin gene expression in the male rat anterior pituitary gland, whilst Y<sub>1</sub> receptor agonists had no effect. The administration of NPY to anterior pituitary cells appears to inhibit prolactin secretion, possibly by amplifying the action of dopamine (Wang et al., 1996), although an earlier group found no *in vitro* effect of NPY on prolactin secretion in bovine pituitary cells (Chao et al., 1987).

### *Neuropeptide Y and gonadotropins*

Central and intravenous injection in animals (Allen et al., 1985; Sahu et al., 1987) and intravenous infusion in humans (Watanobe et al., 1994) causes increased LH, partly by increasing the pituitary response to LHRH (Crowley et al., 1985; Bauer-Dantoin et al., 1991; Denniston et al., 2003). Intravenous NPY administration causes a greater than two-fold elevation in LH secretion (Rodriguez-Sierra et al., 1987) although this effect is not *in vitro* (Chao et al., 1987; Rodriguez-Sierra et al., 1987). Bauer-Dantoin et al. (1992) suggest that NPY only exerts its stimulatory effect on LH release in the presence of adequate concentrations of oestrogen and progesterone and Sahu et al. (1995) demonstrated that NPY mRNA expression increases before the preovulatory LH surge. That NPY plays an important role in the preovulatory LH surge is evidenced by increased hypothalamic NPY levels in young, but not middle aged rats during the LH surge (Sahu and Kalra, 1998).

#### *Neuropeptide Y and thyrotrophin*

Thyroidectomy in the rat is associated with elevation in anterior pituitary NPY (Jones et al., 1994). Intracerebroventricular injection of NPY suppresses circulating T3 and T4 (Fekete et al., 2001) and this inhibitory effect has been shown to involve Y<sub>1</sub> and Y<sub>5</sub> receptors. It is believed that the inhibitory effect of NPY on the thyroid axis is particularly important during fasting (Fekete et al., 2001).

#### *Neuropeptide Y and adrenocorticotrophin*

Intracerebroventricular (Brooks et al., 1994) and intravenous administration (Inoue et al., 1989) of NPY stimulates ACTH and cortisol production in a variety of species. Synaptic contacts exist between NPY- and ACTH-immunoreactive neurones in the hypothalamus (Csiffary et al., 1990) suggesting a functional role for neuropeptide Y in this axis. The stimulatory effect of NPY is inhibited by CRH antagonists (Inoue et al., 1990), suggesting that its effect on ACTH is partly mediated via CRH. This is further supported by the finding that centrally administered NPY increases CRH mRNA in the hypothalamus (Suda et al., 1993). *In vitro* NPY administration to pituitary cells has no effect on ACTH release, suggesting its effect on pituitary corticotrophs is not direct (Small et al., 1998). The only study investigating the intravenous effect of NPY on humans demonstrated an inhibitory effect on nocturnal ACTH and cortisol levels (Antonijevic et al., 2000).

*Neuropeptide Y and pituitary tumours*

Grouzman et al. (1988) demonstrated NPY-immunoreactivity in 33% of human pituitary adenomas, compared with 12% of non-tumorous pituitaries but there was no correlation with tumour activity. Knerr et al. (2001) also demonstrated NPY mRNA in pituitary adenomas. A summary of the effects of NPY on the hypothalamo-pituitary axis is shown in Table 8.

**Table 8. Effects of NPY on the HP axis**

<b>HP axis</b>	<b>Method of administration</b>	<b>Effect</b>	<b>Reference</b>
<b>GH</b>	Central injection	↑↓	McDonald et al., 1985; Thomas et al., 1999; Morrison et al., 2003
	iv infusion (acromegaly)	↑↓	Watanobe et al., 1997
	iv infusion (prolactinoma)	↑	Watanobe et al., 1996
	<i>in vitro</i>	↑	McDonald et al., 1985
	<i>in vitro</i> (somatotroph tumour)	↓	Adams et al., 1987
<b>Prolactin</b>	Central injection (low dose)	↑	Chao et al., 1987
	Central injection (high dose)	↓	Chao et al., 1987
	<i>in vitro</i>	↓	Chao et al., 1987; Wang et al., 1996
<b>LH</b>	Central injection	↑	Allen et al., 1985
	iv infusion (including humans)	↑	Rodriguez-Sierra et al., 1987; Bauer-Dantoin et al., 1991; Bauer-Dantoin et al., 1992; Watanobe et al., 1994
<b>TSH</b>	Central injection	↓	Fekete et al., 2001
<b>ACTH</b>	Central injection	↑	Brooks et al., 1994; Wahlested et al., 1987
	iv infusion	↑	Inoue et al., 1989
	iv infusion (humans)	↓	Antonijevic et al., 2000

**1.13.2 Neuropeptide Y and headache**

NPY plays an important role in the sympathetic innervation of the cerebral vasculature (Edvinsson et al., 1983). NPY-immunoreactive varicose nerve fibres have been demonstrated in cerebral arteries (Edvinsson et al., 1983), cortical veins

(Edvinsson et al., 1984) and free within the dura mater (Keller and Marfurt, 1991). Denervation experiments have shown that most of the sympathetic fibres arise from the ipsilateral superior cervical ganglion (Uddman et al., 1989). Radioimmunoassay studies have also demonstrated the presence of NPY in cerebral arteries, and the addition of NPY to cerebral arteries causes vasoconstriction in a dose-dependent manner (Uddman et al., 1989).

Goadsby et al. (1990) did not find elevated NPY levels during a migraine attack, although Gallai et al. (1994) demonstrated a significant rise in serum NPY during migraine with and without aura in juveniles. NPY levels are not elevated during a cluster attack (Goadsby and Edvinsson, 1994) or in patients with chronic tension type headache (Ashina et al., 1999).

#### **1.14 Vasoactive Intestinal Polypeptide**

VIP is a 28-amino acid peptide that acts on G protein-coupled receptors that activate adenylate cyclase and phospholipase cascades. Three VIP receptors have been cloned that share many biological properties to pituitary adenylate cyclase activating polypeptide (PACAP) receptors, and they are named PACAP/ VIP receptors (PVRs; Nussdorfer and Malendowicz, 1998). VIP and PVRs are widely distributed within the central and peripheral nervous systems and are believed to have a variety of biological actions (Dickinson and Fleetwood-Walker, 1999). VIP-immunoreactive fibres have been localised in laminae I and II of spinal cord dorsal horn, and mRNA expression has been demonstrated in dorsal root ganglion neurones, implicating this peptide in nociception (Noguchi et al., 1989). Intrathecal application of VIP at the spinal cord level has been shown to facilitate nociceptive reflexes (Xu and Wiesenfeld-Hallin, 1991) and the iontophoretic application of VIP causes a marked excitation of rat trigeminal caudalis neurones as well as intact rat (Dickinson et al., 1999) and cat (Jeftinija et al., 1982) spinal cord neurones.

##### **1.14.1 VIP and the pituitary**

VIP has been isolated in high concentrations in both the hypothalamus and the anterior pituitary gland (Lam, 1991). It is a neurotransmitter and neuromodulator within the hypothalamo-pituitary axis, and has a paracrine effect within the pituitary gland itself (Lam, 1991). VIP-containing neurones are present in the suprachiasmatic

nucleus of the hypothalamus as well as the periventricular and paraventricular nuclei (Dai et al., 1997), and VIP immunoreactivity has been demonstrated in the anterior pituitary of several mammalian species (Fahrenkrug and Schaffalitzky de Muckadell, 1978; Besson et al., 1979; Koves et al., 1990; Lam et al., 1990; Carrillo and Dluzen, 1993).

#### *VIP and growth hormone*

VIP causes a marked increase in growth hormone release when added to growth-hormone secreting pituitary tumour cells *in vitro* (Matsushita et al., 1981; Chihara et al., 1982; Fazekas et al., 2000). The stimulatory effect of VIP in acromegaly has also been confirmed *in vivo* (Chihara et al., 1984; Kato et al., 1984).

#### *VIP and prolactin*

VIP stimulates prolactin *in vitro* (Gourdji et al., 1979; Nicosia et al., 1980; Rotsztein et al., 1980; Matsushita et al., 1983) *in vivo* in humans (Bataille et al., 1981; Falsetti et al., 1988; Yiangou et al., 1988) which does not appear to be via dopaminergic inhibitory mechanisms (Tater et al., 1983; Haisenleder et al., 1988). Centrally injected VIP into the hypothalamus stimulates both prolactin secretion (Akema et al., 1988) and mRNA production (Bredow et al., 1994; Di et al., 1997) whilst infusion of anti-VIP serum into the portal vein in rats suppresses serotonin-induced prolactin secretion (Shimatsu et al., 1984). VIP enhances the secretion of prolactin in pituitary tumours both *in vitro* (Kato et al., 1984; Prysor-Jones et al., 1984) and *in vivo* (Kato et al., 1984). Fazekas et al. (2000) demonstrated that addition of VIP to growth hormone-secreting pituitary tumours and prolactinomas caused increased prolactin release and an increase in the presence of secretory granules within the adenoma cells.

#### *VIP and gonadotropins*

*In vitro* treatment of pituitary tissue with VIP does not affect the release of LH or FSH (Enjalbert et al., 1980; Rotsztein et al., 1980; Sawangjaroen et al., 1997) although micro-injection of VIP into the hypothalamus and the third ventricle inhibits LH pulses (Akema et al., 1988; Weick et al., 1992). Falsetti et al. (1988) demonstrated that the intravenous VIP administration in women does not modify serum LH or FSH, although Hammond et al. (1993) showed that it may modulate the responsiveness of

the gonadotroph to LHRH. It has been suggested that VIP plays a modulatory, rather than 'deterministic', role in the regulation of LH secretion (Weick and Stobie, 1995).

#### *VIP and thyrotrophin*

Central VIP injection may stimulate the hypothalamo-pituitary-thyroid axis in rats (Mitsuma et al., 1984) although *in vitro* treatment of pituitary tissue with VIP (Baranowska et al., 1999) and intravenous administration in humans (Falsetti et al., 1988) does alter serum TSH levels. Thyroidectomy increases anterior pituitary VIP (Michalkiewicz et al., 1987; Buhl et al., 1995) suggesting that some hypothalamic factor is involved in mediating the effect of hypothyroidism on pituitary VIP expression and secretion.

#### *VIP and adrenocorticotrophin*

Central VIP injection increases ACTH secretion (Itoh et al., 1982; Itoh and Hirota, 1983; Alexander and Sander, 1994). In humans, intravenous VIP infusion does not alter serum ACTH in healthy volunteers (Ambrosi et al., 1987; Watanobe and Tamura, 1994; Chiodera et al., 1996). VIP elevates ACTH in patients with ACTH-secreting pituitary tumours (Ambrosi et al., 1987; Watanobe and Tamura, 1994) suggesting that neoplastic transformation of pituitary cells may be associated with their capacity to express PACAP/ VIP receptors (PVRs). *In vitro* investigations have shown no effect of VIP or peptide histidine-methionine (PHM, a compound derived from a VIP precursor molecule with similar biological actions to VIP) on ACTH secretion in normal perfused pituitary cells or incubated hemipituitaries of rats and mice (Nicholson et al., 1984; Tilders et al., 1984; Vigh and Schally, 1984) although a stimulatory effect is observed in the presence of CRH (Tilders et al., 1984; Vigh and Schally, 1984). Experiments on pituitary adenoma tissue support the finding that VIP has a stimulatory effect on ACTH secretion in corticotrope pituitary tumour cells compared to the normal pituitary gland (White et al., 1982; Westendorf et al., 1983)

#### *VIP and pituitary tumours*

PVRs are differentially expressed in pituitary adenomas, suggesting that abnormal regulation of VIP-related pathways are important in the pathogenesis of certain pituitary tumours (Oka et al., 1998). Hsu et al. (1989) demonstrated strong VIP

immuno-reactivity in human pituitary tumours. They found significant quantities of VIP in 16 of 17 prolactinomas, 12 of 14 growth hormone-secreting tumours, 4 of 12 ACTH-secreting tumours and 14 of 18 non-functioning adenomas. Double immunolabelling demonstrated VIP-immunoreactivity in many lactotrophs, thyrotrophs, corticotrophs and the occasional gonadotroph (Hsu et al., 1989). A summary of the effects of VIP on the hypothalamo-pituitary axis is shown in Table 9.

**Table 9. Effects of VIP on the HP axis**

<b>HP Axis</b>	<b>VIP administration</b>	<b>Effect</b>	<b>Reference</b>
<b>GH</b>	in vitro (acromegaly)	↑	Matsushita et al., 1981; Chihara et al., 1982; Fazekas et al., 2000
	iv infusion (acromegaly)	↑	Chihara et al., 1984; Kato et al., 1984
<b>Prolactin</b>	in vitro	↑	Gourdji et al., 1979; Nicosia et al., 1980; Rotsztejn et al., 1980
	in vitro (acromegaly, prolactinoma)	↑	Pryor-Jones et al., 1984; Kato et al., 1984
	Central injection	↑	Akema et al., 1988; Bredow et al., 1994; Di et al., 1997
	iv infusion (healthy volunteers)	↑	Bataille et al., 1981; Falsetti et al., 1988; Yiangou et al., 1988
	iv infusion (acromegaly, prolactinoma)	↑	Kato et al., 1984
<b>LH/FSH</b>	in vitro	→	Enjalbert et al., 1980; Rotsztejn et al., 1980; Sawangjaroen et al., 1997
	Central injection	↓	Akema et al., 1988; Weick et al., 1992
	iv humans	→↑	Falsetti et al., 1988; Hammond et al., 1993
<b>TSH</b>	Central injection	↑	Mitsuma et al., 1984
	in vitro	→	Baranowska et al., 1999
	iv humans	→	Falsetti et al., 1988
<b>ACTH</b>	Central injection	↑	Itoh et al., 1982; Itoh et al., 1983
	iv humans (healthy volunteers)	→	Ambrosi et al., 1987; Chiodera et al., 1996; Watanobe et al., 1994
	iv humans (Cushings disease)	↑	Ambrosi et al., 1987; Watanobe et al., 1994
	in vitro	→	Nicholson et al., 1984; Tilders et al., 1984; Vigh et al., 1984
	in vitro (with CRH)	↑	Tilders et al., 1984; Vigh et al., 1984
	In vitro (Cushings)	↑	Westendorf et al., 1983; White et al., 1982



### 1.14.2 VIP and headache

VIP-immunoreactive nerve fibres are present in the parasympathetic innervation of the cerebral arteries, and these fibres may originate from the pterygopalatine, otic and internal carotid ganglia (Gibbins et al., 1984; Suzuki et al., 1988; Hara et al., 1989). Radioimmuno-assay studies confirm the presence of VIP in human cerebral and middle meningeal arteries (Edvinsson et al., 1987) and pharmacological studies show VIP to be a potent vasodilator of the latter (Edvinsson et al., 1987). VIP-immunoreactive fibres have been demonstrated in cerebral venous sinuses and free within the dura mater (Keller and Marfurt, 1991). VIP levels are elevated following superior sagittal sinus stimulation (Zagami et al., 1990) as a result of trigeminal-autonomic activation. Neither peripheral nor jugular VIP levels are altered during migraine with or without aura in humans (Blegvad et al., 1986; Goadsby et al., 1990) and Nicolodi et al. (1990) demonstrated reduced salivary VIP levels in migraine. Conversely, salivary (Nicolodi and Del Bianco, 1990) and external jugular (Goadsby and Edvinsson, 1994) VIP levels are elevated during cluster headache and chronic paroxysmal hemicrania attacks (Goadsby and Edvinsson, 1996) with resolution to normal levels after the attacks are finished.

#### Summary

There are several neuropeptides that may play an important role in headache pathophysiology (Table 10) that also appear to have paracrine activity within the hypothalamo-pituitary axis, and are differentially expressed in pituitary tumours. It is conceivable that the secretion of these peptides may be involved in pituitary-tumour associated headache.

**Table 10. Neuropeptide changes in primary headache**

	<b>Migraine</b>	<b>Cluster Headache</b>	<b>Chronic Tension Type Headache</b>
<b>CGRP</b>	↑	↑	→ / ↑ *
<b>Substance P</b>	-	-	-
<b>Neuropeptide Y</b>	- / ↑	-	-
<b>VIP</b>	- / ↓	↑	-

\* CGRP increased with throbbing tension-type headache only

## 1.15 Somatostatin and Headache

The final part of this introduction is a review of the literature regarding the role of somatostatin and headache. Somatostatin was initially isolated as a 14-amino acid peptide that reduced the release of growth hormone from the pituitary gland (Brazeau et al., 1973). In addition to its neuroendocrine role, somatostatin has subsequently been found to have diverse neurophysiological effects (Epelbaum et al., 1994; Schindler et al., 1996). This discussion will be limited to its role in nociception, primary headache and pituitary-associated headache.

### 1.15.1 Non-Clinical Data

Somatostatin is translated as a pre-propeptide, of which two predominant biologically active metabolites have been found, somatostatin-14 and the amino-terminally extended form, somatostatin-28. A family of five somatostatin receptor genes have been cloned, termed *sst*<sub>1-5</sub>, and based on pharmacological properties and sequence analysis, the receptors have been divided into two groups (Table 11). The somatostatin receptor-1 group consists of *sst*<sub>2</sub>, *sst*<sub>3</sub> and *sst*<sub>5</sub>, and the somatostatin receptor-2 group consists of *sst*<sub>1</sub> and *sst*<sub>4</sub> (Hoyer et al., 1995). In mice and rats, the *sst*<sub>2</sub> receptor is found as two splice variants, *sst*<sub>2(a)</sub> and *sst*<sub>2(b)</sub>, which differ in length and composition of their respective carboxy-terminal (Hoyer et al., 1995). The somatostatin receptors are G-protein coupled seven-transmembrane receptors, which interact with a wide range of downstream signalling targets that includes cAMP (Selmer et al., 2000).

**Table 11. Classification of *sst* receptors**

<b>Group 1</b>	<b>Group 2</b>
<i>sst</i> <sub>2</sub>	<i>sst</i> <sub>1</sub>
<i>sst</i> <sub>3</sub>	<i>sst</i> <sub>4</sub>
<i>sst</i> <sub>5</sub>	

Somatostatin is located within sensory ganglia (Smith et al., 1993) as well as in the trigeminal ganglion and the trigeminal nucleus caudalis (Alvarez and Priestley, 1990; Lazarov and Chouchkov, 1990; Del Fiacco and Quartu, 1994; Yin, 1995).

Somatostatin is also localised within the substantia gelatinosa of the dorsal horn of the spinal trigeminal nucleus (Johansson et al., 1984; Kiyama and Emson, 1990; Mengod et al., 1992) strongly suggesting that this peptide has a role in trigeminal nociception.

It is believed that the  $sst_{2(a)}$  receptor is particularly important in somatostatin-associated analgesia. Humphrey's group in Cambridge has done much of the work on the immuno-localisation of somatostatin receptors, demonstrating  $sst_{2a}$  receptors in the periaqueductal gray and dorsal horn neurones (Schindler et al., 1997; Schindler et al., 1998).

The precise mechanism of the analgesic action of somatostatin has not been elucidated, but there is animal evidence that its actions are probably central. The peptide nature of somatostatin renders it vulnerable to breakdown by endogenous peptidases, and its short half-life of several minutes (Harris, 1994) has led to the development of longer acting analogues, such as octreotide. Intrathecal and subcutaneous injection of somatostatin and octreotide respectively, have been shown to inhibit the firing of dorsal horn neurones (Chapman and Dickenson, 1992) and direct micro-injection of somatostatin into the periaqueductal gray of the cat has been shown to produce inhibition of heat-induced electrophysiological recordings in the spinal cord (Helmchen et al., 1995). Micro-injection of somatostatin into the caudate putamen has also been shown to be anti-nociceptive (Tashev et al., 2001). Song et al. (2002) showed that  $sst_{2(a)}$  receptors are preferentially involved in thermo-nociception but not mechano-nociception and Su et al. (2001) demonstrated that octreotide has no peripheral modulatory action in visceral nociception, suggesting that its site of action is central rather than peripheral. Heyles et al. (2001) demonstrated that somatostatin analogues inhibit mustard oil-evoked neurogenic plasma extravasation and inhibition of the release of substance P and CGRP in response to electrical stimulation which suggests an anti-inflammatory role for somatostatin. Earlier studies have also supported an anti-inflammatory role for somatostatin. For example, Matsubara et al. (1992) demonstrated that octreotide is able to block plasma protein leakage within dura mater.

Octreotide has been shown to inhibit substance P release from sensory nerve endings and to inhibit substance P-induced vasodilation (Gazelius et al., 1981). This has led certain groups to believe that the analgesic action of somatostatin analogues, particularly in relation to headache, is via direct vascular mechanisms rather than a central action. Sicuteri et al. (1984) showed that when the dorsal hand vein is locally injected with native somatostatin, it spasms visibly. This study showed that

tachyphylaxis develops following 2-4 injections of somatostatin, and this is reversed by the local injection of naloxone. That somatostatin may have a direct effect on blood vessels is evidenced by the finding of its receptors on human vasculature, sst<sub>1</sub> having a significantly higher level of expression than sst<sub>2</sub> and sst<sub>4</sub> (Curtis et al., 2000).

Octreotide is known to have poor penetration of the blood-brain barrier (Kitazawa et al., 1998; Schmidt et al., 1998) due to its non-lipophilic properties, which has been used by some as circumstantial evidence of a peripheral action of octreotide in headache (Kemper et al., 2000). Kemper et al. (2000) looked at capsaicin-induced c-Fos expression in an animal model of trigeminal nociception. This group showed that the intracisternal administration of octreotide does not alter capsaicin-induced c-Fos expression and they suggest that this argues against a central role for somatostatin receptors in the processing of trigeminovascular signals. Conversely, Berieter (1997) demonstrated that the intracerebroventricular administration of octreotide reduced c-Fos expression in the trigeminal subnucleus in response to corneal stimulation in the rat. Furthermore, Berieter (1997) demonstrated that the co-administration of sub-therapeutic doses of morphine potentiated this analgesic effect, suggesting that somatostatin and opiates may have interacting or overlapping properties. Somatostatin analogues have been previously shown to have affinity for opiate receptors (Rezek et al., 1978; Maurer et al., 1982; Pelton et al., 1985; Pelton et al., 1985) and there is evidence to suggest that somatostatin and opiate receptors may heterodimerise (Pfeiffer et al., 2002). Samsam et al. (2002) electrically stimulated the trigeminal ganglia of rats and studied the subsequent immuno-reactivity for methionine enkephalin, neurotensin and somatostatin. They observed a decrease in immunoreactivity for all three peptides ipsilateral to the side of stimulation, and they suggest that this may be evidence for their potential role as antinociceptive agents during trigeminal activation.

At the cellular electrophysiological level, somatostatin has been shown to have both excitatory as well as inhibitory actions on neurones (Dodd and Kelly, 1978; Pittman and Siggins, 1981; Mueller et al., 1986). Somatostatin appears to have a reciprocal relationship with glutamate (Hathway et al., 2001). Glutamate is believed to play an important role in the transmission of trigemino-vascular nociceptive information (Storer and Goadsby, 1999) and the inhibitory action of somatostatin on trigeminal

neuronal activity may be partly explained by its modulation of glutamate-mediated nociceptive signals (Gardette et al., 1995; Lanneau et al., 1998; Peineau et al., 2003). There is also evidence of an interaction between somatostatin and  $\gamma$ -aminobutyric acid (GABA). The GABA<sub>A</sub> receptor appears to be involved in pain transmission within the trigeminal system (Storer et al., 2001) and somatostatin has been shown to have a functional interaction with the GABA<sub>A</sub> receptor (Vincens et al., 1998) making it possible that the analgesic effect of somatostatin involves enhancement in the activity of this receptor.

### **1.15.2 Clinical Data**

#### *Somatostatin and Peripheral Pain*

There is evidence in the clinical literature that somatostatin and its analogues may be potent analgesic agents in the management of a variety of conditions characterised by acute and chronic pain. Chrubasik et al. (1984) first reported in the Lancet a series of eight patients with terminal metastatic cancer pain who received both intrathecal and epidural infusions of native somatostatin. In this open label study, somatostatin was found to be an effective analgesic agent when administered in the epidural space, and the effect was not reversed with naloxone which suggests a non-opioidergic mechanism of action. The same group found continuous epidural infusions of somatostatin to be effective in providing complete post-operative pain relief in eight patients who had undergone abdominal surgery. In two of these patients, epidural somatostatin also provided adequate intraoperative analgesia (Chrubasik et al., 1985). In a larger placebo controlled study of 40 patients undergoing abdominal surgery, epidural somatostatin was found to be significantly better than placebo for pain relief and the need for additional analgesia was higher in the placebo group (Taura et al., 1994). Fioravanti et al. (1995) injected somatostatin intra-articularly in 41 patients with rheumatoid arthritis in an open label study, and found a significant reduction in pain and markers of inflammation.

Octreotide, a somatostatin analogue that acts predominantly on sst<sub>2</sub> and sst<sub>5</sub> (Patel and Srikant, 1994) has been shown to be as equally useful intrathecally as native somatostatin in the management of severe cancer pain (Penn et al., 1992). Octreotide is also known to be effective in the treatment of musculoskeletal symptoms associated with carcinoid syndrome (Smith et al., 1990), the latter tumours being known to

secrete CGRP (Takami et al., 1990), substance P (Emson et al., 1984), neuropeptide Y (Waeber et al., 1995) and VIP (Virgolini et al., 1994).

### *Somatostatin and Primary Headache*

There have been few clinical trials studying the usefulness of somatostatin and its analogues in the management of primary headache syndromes, although the preliminary evidence suggests that they may be effective agents for both cluster headache and migraine. Sicuteri et al. (1984) published one of two currently available studies concerning the effect of somatostatin in cluster headache. This group investigated eight males with cluster headache and hospitalised them during their cluster bout. All patients were withdrawn from preventative treatment 4 days prior to the study. Patients received 3 double-blind treatments of somatostatin, ergotamine and placebo during a cluster attack. Pain intensity was assessed by a visual analogue scale every 10 minutes, and the 'area of pain' was studied for each treatment. The study found somatostatin to reduce the pain area by 32%, whilst ergotamine reduced this by 45%. The differences between somatostatin and ergotamine were not significant, and both were significantly more effective than placebo. In the discussion section of the paper, Sicuteri et al. (1984) acknowledge the difficulty in studying a large number of patients with cluster headache and concede that 'strictly controlled clinical studies (are) still required'. The second randomised study was a non-placebo controlled double blind study comparing the efficacy of somatostatin with ergotamine in cluster headache (Geppetti et al., 1985). Five patients were treated for three attacks by each drug. Subcutaneous somatostatin and ergotamine were equally beneficial as regards effects on maximal pain intensity and pain area, but somatostatin was less effective in reducing the duration of pain. This limited evidence of the beneficial effect of somatostatin in cluster headache needs to be explored further in properly controlled and adequately powered studies.

The only available study concerning the effect of somatostatin receptor activation on migraine was conducted by Kapicioglu et al. (1997). In this double-blind parallel study, octreotide (100µg) was given subcutaneously to 17 patients with migraine with and without aura, and placebo was given to 12 matched patients. At 2, 4 and 6 hours, headache relief, defined as reduction in severity from grade 3 or 2 (severe or

moderate) to 1 or 0 (mild or none), was found to be significantly better in patients taking octreotide than placebo. By 6 hours, headache relief was experienced in 77% of octreotide-treated patients, and 25% of the placebo-treated group. The authors' rationale for the study was that octreotide 'causes a marked reduction in the concentration of substance P and VIP...(which) may be involved in the pathogenesis of migraine attacks'. They conclude that octreotide 'is very effective and well tolerated for the treatment of a migraine attack, but additional studies will be necessary to verify these conclusions'.

Since that study, no other groups have followed up this potentially interesting observation.

#### *Somatostatin and pituitary tumour-associated headache*

Although the reports on the use of somatostatin analogues directly aimed at the treatment of primary headaches are not numerous, there are an increasing number of clinical studies describing the effectiveness of these agents in the treatment of the headache associated with acromegaly.

The two somatostatin analogues most commonly used in the treatment of acromegaly are octreotide and lanreotide. Large-scale placebo-controlled trials have shown that short-acting octreotide can reduce headache in approximately 75-80% of patients with acromegaly (Ezzat et al., 1992; Newman et al., 1995; Newman et al., 1998) and the use of monthly slow-release octreotide has been shown to give a reduction in symptoms after the sixth injection (Lancranjan and Atkinson, 1999). The prospective trials of lanreotide therapy have also shown a significant reduction in headache in addition to the other symptoms of acromegaly, including fatigue, perspiration, joint pains and parasthesia (Cannavo et al., 2000; Cannavo et al., 2001) although a subgroup of patients is recognised who prefer to be transferred to octreotide therapy because only the latter agent significantly ameliorates headache (Cannavo et al., 2000). Our own Case 3 (Appendix; Levy et al., 2003b) is a good example of such a patient, and suggests that there is a different mechanism for the analgesic effect of octreotide from its growth hormone lowering action.

Octreotide and lanreotide predominantly bind to sst<sub>2</sub> and sst<sub>5</sub> (Patel and Srikant, 1994). Functional pituitary tumours have been shown to express sst<sub>1</sub>, sst<sub>2</sub>, sst<sub>3</sub> and sst<sub>5</sub> (Schaer

et al., 1997; Shimon and Melmed, 1997). Using tumour cell cultures, it has been shown that both  $sst_2$  and  $sst_5$  are involved in the regulation of growth hormone release from growth hormone-secreting pituitary tumours, whereas  $sst_5$  appears to be exclusively involved in the regulation of prolactin secretion from prolactinoma cells (Shimon et al., 1997). Octreotide has significantly better binding to  $sst_3$  than lanreotide (Patel and Srikant, 1994) and it is possible that the subgroup of patients who respond only to octreotide in terms of headache, but both octreotide and lanreotide in terms of growth hormone, do so because their tumours express  $sst_3$ , coupled to suppression of the pain pathway, and  $sst_5$  receptors, coupled to the control of GH release (Levy et al., 2003b). This hypothesis requires further study.

Although somatostatin analogues are predominantly used in clinical practice for their ability to lower growth hormone in acromegaly, there is evidence that they also have an analgesic effect in non-growth hormone secreting pituitary tumours, including prolactinomas (Williams et al., 1986), non-functioning adenomas (Turpin et al., 1991) and TSH-secreting tumours (Yoenem et al., 1999). It is possible that the analgesic action of octreotide in these cases is similarly related to the somatostatin receptor status of the tumour.

An alternative explanation for the differential analgesic effect of octreotide and lanreotide in headache is related to central anti-nociceptive mechanisms rather than direct effects on the pituitary gland. As discussed previously, the periaqueductal gray, hypothalamus and trigeminal nucleus are known to possess  $sst_{2a}$  receptors (Schindler et al., 1997) and represent potential targets for the analgesic action of somatostatin analogues. However, octreotide and lanreotide both exhibit limited permeability to the blood-brain barrier (Gillespie et al., 1998; Fricker et al., 2002) and differences in action seem more likely to be related to pharmacodynamics than pharmacokinetics. This situation is analogous to the triptans, which also have the combination of apparent poor central nervous system accessibility (De Vries et al., 1999) and undoubted efficacy in primary headache (Ferrari et al., 2002). It has been suggested that there may be an altered vascular permeability during activation of the trigeminovascular system when compared to the resting state to account for the paradox between poor permeability of the blood brain barrier and apparent efficacy



during headache (Harper et al., 1977; Alvarez-Cermeno et al., 1986; Kaube et al., 1993).

Finally, it is of interest that pegvisomant, a new agent that has been developed for the treatment for acromegaly, appears to be less helpful for headache than somatostatin analogues. Unlike somatostatin analogues, pegvisomant is a peripheral growth hormone-receptor blocker and does not act centrally on the hypothalamo-pituitary axis. This agent is very effective in reducing all the typical symptoms of acromegaly apart from headache (van der Lely et al., 2001) which supports a central and specific analgesic action of somatostatin analogues, separate from the ability to improve other symptoms related to growth hormone excess.

#### *Somatostatin and Headache Induction*

Although the majority of the literature regarding somatostatin as regards pain concerns its anti-nociceptive properties, there is evidence that repeated and prolonged exposure to somatostatin may be pro-nociceptive. There have been two reports of rebound headache and dependency to octreotide (Popovic et al., 1988; May et al., 1994) suggesting that there is an up-regulation of trigeminal nociceptive pathways after excessive exposure and subsequent withdrawal of this agent. Octreotide dependency is also described in our own Case 5 (Appendix; Levy and Goadsby 2004). Octreotide has been shown to trigger cluster headache attacks in certain patients (Otsuka et al., 1998) in stark contrast to other reports of the abortive effects of somatostatin in this condition (Sicuteri et al., 1984; Geppetti et al., 1985). The administration of native somatostatin to healthy individuals causes suppression of growth hormone, which rebounds when it is discontinued, and endocrinologists have observed that patients commonly experience headache after somatostatin withdrawal during metabolic studies (Lightman, unpublished observations). Despite this observation, there have been no previous studies directly observing the effects of somatostatin infusion and withdrawal on headache, and to correlate this with neuroendocrine changes. One of the problems of studying primary headache syndromes such as migraine and cluster headache in clinical research, is the episodic nature of these conditions, and to have a reliable way of triggering headache in these conditions is very attractive. Nitroglycerin (Thomsen et al., 1994) and histamine (Krabbe and Olesen, 1980) have both been used to induce migraine and cluster

headache attacks. Patients with migraine develop symptoms usually several hours after exposure to nitroglycerin (Iversen, 2001), whilst cluster patients (during a cluster period) may have an attack within minutes (Ekbom, 1968). A problem with these existing models is that the most reliable and best studied model, that of nitroglycerin-triggering, is complicated by vasodilatation (Thomsen et al., 1994) and dissecting the non-vascular mechanisms in these headache types is a considerable challenge. It would thus be desirable to have a non-vasoactive method for triggering migraine or cluster headache, or both. Somatostatin may therefore represent a compound that may be of use in the further study and potential triggering of primary headache.

In summary, this introduction has outlined the currently available evidence to support the view that hypothalamo-pituitary axis and somatostatin are relevant to the study of headache. The clinical features and classification of the headache phenotypes encountered in this thesis have been discussed. The range of headache syndromes previously described in association with pituitary tumours have been presented, and the proposed mechanisms of headache in pituitary tumours have been reviewed, with particular emphasis on certain vasoactive neuropeptides that appear to play a role both in the pituitary gland and headache pathophysiology. Finally, the relevance of somatostatin to primary and pituitary-associated headache has been discussed.

## **1.16 Thesis Aims**

The aims of the thesis are as follows:

- (i.) To determine the importance of structural factors in pituitary tumour-associated headache (Chapter 2)
- (ii.) To determine the importance of certain neuropeptides in pituitary tumour-associated headache (Chapter 3)
- (iii.) To describe the headache phenotypes and treatment characteristics in a cohort of patients with pituitary tumour-associated headache (Chapter 4)
- (iv.) To test the hypothesis that somatostatin withdrawal may be used as a means of triggering headache (Chapter 5)
- (v.) To investigate the usefulness of octreotide in migraine and cluster headache (Chapter 6)

**2. Pituitary tumours and headache: structural factors**

## **2.1. Study 1: the relationship between pituitary tumour volume, cavernous sinus invasion and headache**

Levy MJ, Jäger HR, Powell M, Matharu MS, Meeran K, Goadsby PJ

Abstract ref: Cephalalgia 2002; 22: 592

Paper ref: Archives of Neurology 2004; 61(5): 721-5

### **2.1.1 Abstract**

*Background:* Pituitary tumours are commonly associated with disabling headache. The accepted mechanisms for headache are dural stretch and cavernous sinus invasion.

*Methods:* Sixty-three patients presenting with pituitary tumours were studied prospectively. Headache scores, pituitary tumour volume and extent of cavernous sinus invasion were obtained for each patient.

*Results:* The prevalence of headache was 70%. There was no positive correlation between headache score and pituitary volume ( $r = -0.32$ ,  $P = 0.01$ ). There was no association between cavernous sinus invasion and headache. There was a strong association between pituitary tumour-associated headache and a family history of headache ( $\chi^2 = 8.36$ ,  $P = 0.004$ ).

*Conclusion:* Pituitary tumour-associated headache cannot be just a structural problem. Other factors, such as the presence of a family history of headache and the endocrine activity of the tumour, may be equally important in this phenomenon.

### **2.1.2 Introduction**

As discussed in Section 1.9, it has long been considered that headache is related to tumour size and dural stretch (Forsyth and Posner, 1993; Suwanwela et al., 1994). The explanation for dural stretch as a cause of headache is that the expansion of a pituitary tumour within the sella turcica may stimulate afferent fibres innervating the dura mater. Involvement of the cavernous sinus has also been invoked to explain headache (de Groot & Jameson, 2000) since the sinus contains the ophthalmic branch of the trigeminal nerve and the internal carotid artery, both of which are important structures in head pain and cranial autonomic function respectively.

The numerous reports of a variety of headache phenotypes associated with pituitary tumours, which include severe and intractable migraine (Lee, 1990), cluster headache (Milos et al., 1996; Porta-Etessam et al., 2001), 'trigeminal neuralgia' (Gazioglu, 2000; Gelabert Gonzalez, 1990) and SUNCT (Ferrari et al., 1988; Massiou et al., 2002), have been described in detail in the general introduction. In such cases, conventional preventive and abortive headache management can often prove to be ineffective, yet medical treatment of the pituitary disease can completely resolve the symptoms. The reported cases of micro-prolactinomas presenting with severe headache that resolve immediately with the administration of dopamine agonists (Hartman et al., 1995; Gabrielli et al., 2002) and the impressive analgesic effects of somatostatin analogues in acromegaly (Musolino et al., 1990; Pascual et al., 1991) suggest that biochemical mechanisms may be involved in pituitary tumour-associated headache. Also, the reports of significant headache exacerbations associated with the administration of dopamine-agonists in prolactinomas, in the absence of a change in physical properties of the tumour (Massiou, 2002; Levy 2003), suggest that non-mechanical factors may be involved in the pathophysiology of pituitary tumour-associated headache.

The aim of the study was to systematically look at the relative importance of size and cavernous sinus invasion in pituitary-associated headache to directly test both hypotheses for the cause of headache.

### **2.1.3 Subjects and methods**

Sixty-three consecutive patients (16 male, 47 female; mean age  $44 \pm 14$  years) presenting with pituitary disease from February 2000 to August 2002 were prospectively included in the study. All patients were seen at the same unit (QS) for treatment of newly diagnosed pituitary disease, which included both surgical and medical management options. The study was approved by the Joint Ethical Committee of the National Hospital for Neurology and Neurosurgery and the Institute of Neurology (Study Reference 01/ N022). The following criteria were assessed at presentation:

#### *Headache*

Before commencement of treatment, all patients were interviewed by a trained headache fellow (the candidate). The clinical data collected included the presence or absence of headache, the frequency and severity of symptoms. A retrospective headache score was calculated using the following:

Headache frequency (days/wk) x Headache Duration (hrs/day) x Headache Severity (0-10)

In order to validate this measurement, 22 of the recruited patients were asked to complete prospective headache diaries, in which an hourly headache score was documented for a period of two weeks. These diaries were sent to a separate headache specialist (collaborator Dr MS Matharu) who blindly calculated a mean headache score for each patient. The diary score and the retrospective score were then compared. The presence of a family history of headache was also documented.

#### *Assessment of Pituitary Volume and Cavernous Sinus Invasion*

The structural factors measured in this study were pituitary tumour size and cavernous sinus invasion. In the only previous study investigating headache in pituitary tumours (Abe et al., 1998) pituitary size was estimated using maximum tumour diameter, and objective classification criteria were not described in the assessment of cavernous sinus invasion. I sought to improve on this by determining pituitary volume rather than diameter, and by using objective rather than subjective criteria for cavernous sinus invasion.

The pre-treatment MRI scans were performed on a number of different MRI scanners, all at 1.5T. All examinations included coronal and sagittal T1-weighted spin echo sequences with a maximum slice thickness of 3mm, before and after intravenous administration of a gadolinium-based contrast medium. All MRI scans were assessed by a consultant neuro-radiologist (Dr HR Jäger, Institute of Neurology, London UK) as well as myself. For the assessment of tumour volume it was assumed that pituitary tumours have an ellipsoid shape, in response to a previous study which reported this to be an accurate assessment of pituitary size (Lundin and Pedersen, 1992).

### *Pituitary Volume*

Using Cavalieri's principle, pituitary tumour volume was calculated after performing measurements of tumour diameter in three orthogonal planes (Lundin and Pederson, 1992) using the following equation:

$$\text{Volume} = \frac{4}{3} \pi \cdot \frac{a}{2} \cdot \frac{b}{2} \cdot \frac{c}{2}$$

If the tumour was large and multi-lobed, the tumour volume was assumed to consist of separated ellipses, and the sum of each lobe volume was calculated (Figure 5).

### *Cavernous Sinus Invasion*

Three different parameters were used for the assessment of presence and degree of cavernous sinus involvement, based on previous radiological data (Cottier et al., 2000):

1. Encasement of the internal carotid artery, distinguishing four grades:
  - a. no encasement,
  - b. < 25% encasement
  - c. > 50 - 75% encasement
  - d. > 75 - 100% encasement
2. Crossing of the three lines connecting the cross sections through the distal internal carotid arteries (inter-carotid lines): medial, median and lateral inter-carotid lines.
3. Extension of the tumour into the venous compartments of the cavernous sinus.

Based on a modification of the classification system described by Cottier et al. (2000) tumour extension was divided into the superior, lateral and inferior venous

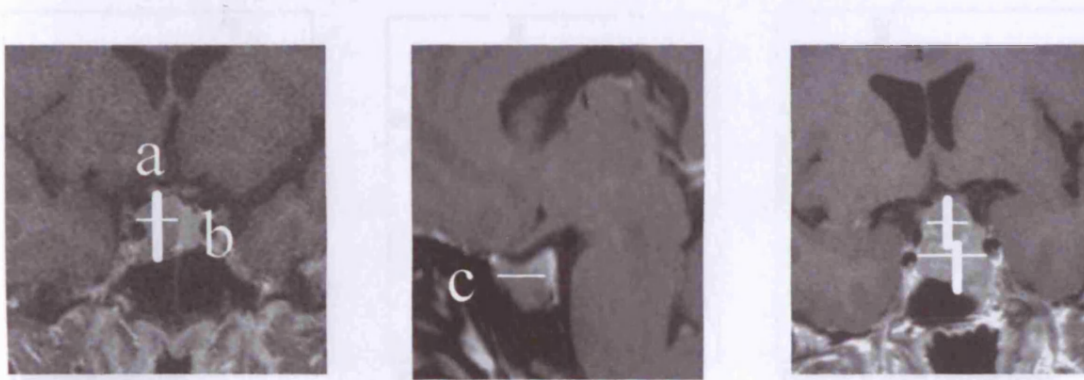


compartments (Figure 6). Because invasion of the medial compartment has not proven to be a significant radiological sign of cavernous sinus invasion (Cottier et al., 2000) this compartment was not included in our classification system. If a normal pituitary gland was observed between the adenoma and the intracavernous internal carotid artery, the cavernous sinus was considered to be free of invasion (Scotti et al., 1988; Knosp et al., 1993). The inferolateral venous and carotid sulcus compartments described by Cottier et al. (2002) were grouped together as the inferior compartment for simplicity (Figure 6). Using previously published data comparing MRI classification with surgical findings (Knosp et al., 1993; Cottier et al., 2000) the following criteria were used to diagnose cavernous sinus invasion (Figure 7):

1. Tumour crosses lateral inter-carotid line
2. Tumour encases >75% internal carotid artery
3. Tumour extends into inferior compartment

Examples of superior, inferior, lateral and absent cavernous sinus invasion are shown in Figure 8.

**Figure 5. Calculation of pituitary volume**

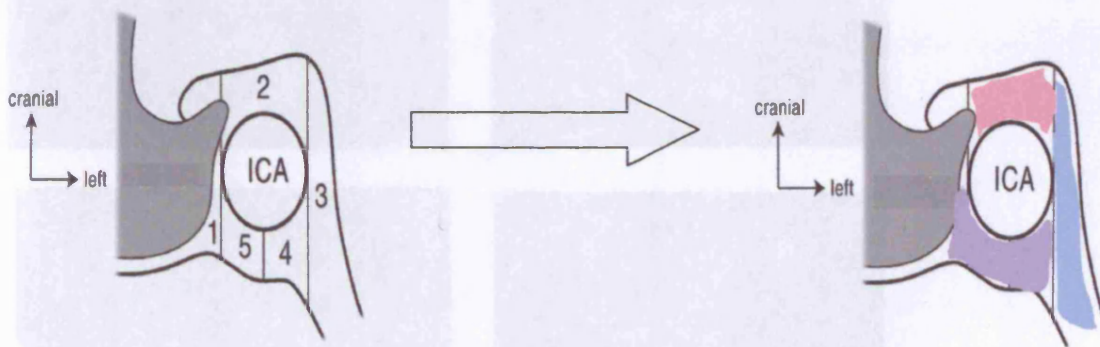


$$\text{Volume} = \frac{4}{3} \pi \cdot \frac{a}{2} \cdot \frac{b}{2} \cdot \frac{c}{2}$$

Multi-lobed tumours are assumed to be the sum of separate ellipses

**Figure 6. Simplification of cavernous sinus invasion into superior, inferior and lateral compartments**

**Figure 6. Simplification of cavernous sinus invasion into superior, inferior and lateral compartments**



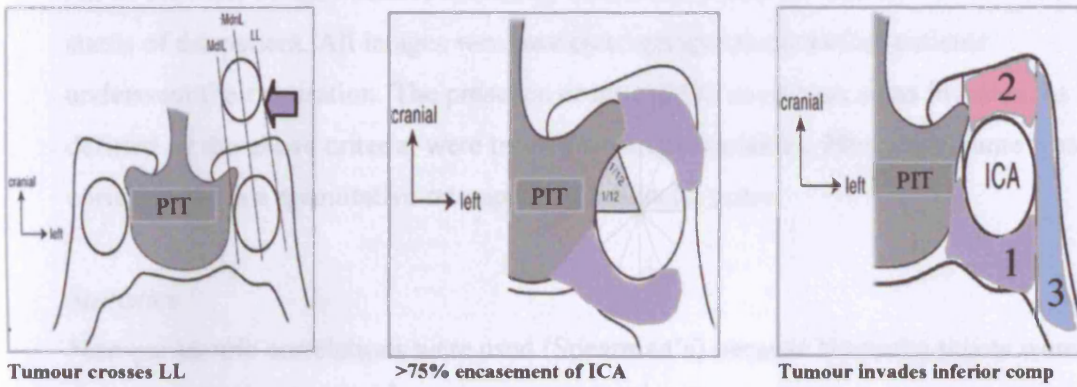
Cottier's classification (2000):

- 1 = medial,
- 2 = superior
- 3 = lateral
- 4 = inferolateral,
- 5 = carotid sulcus

Simplified classification:

- pink = superior
- mauve = inferior
- blue = lateral

**Figure 7. Criteria for cavernous sinus invasion**



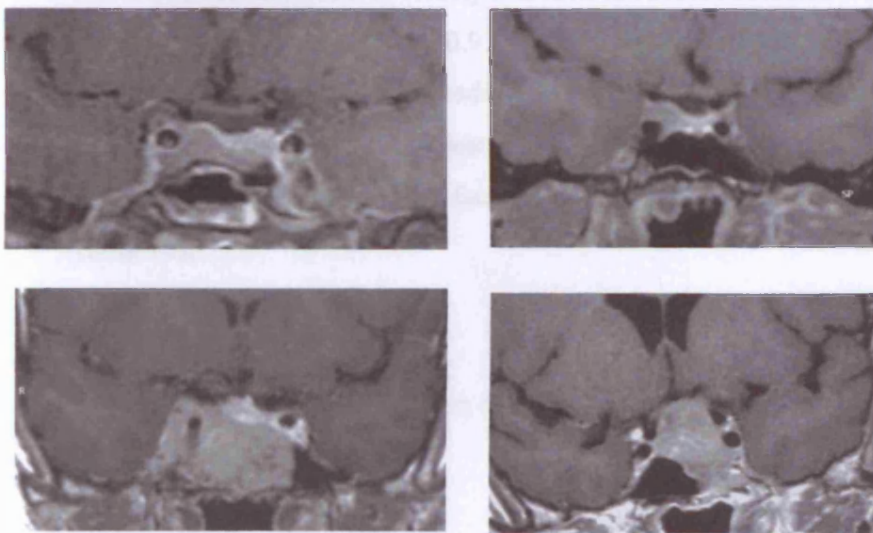
Intercarotid lines: Mdl = medial, MdnL = median, LL = lateral

ICA = internal carotid artery

Cavernous compartments: 1 = inferior, 2 = superior, 3 = lateral

Comp = Compartment

**Figure 8. Examples of cavernous sinus invasion**



Inferior (top left), superior (top right), lateral (bottom left) and no invasion (bottom right)

### *Study design*

All of the MRI images were reviewed by consensus whilst blinded to the headache status of the patient. All images were reviewed prospectively before patients underwent their operation. The presence or absence of cavernous sinus invasion, as defined by the above criteria, were treated as binary variables. Pituitary volume was correlated with a quantitative retrospective headache score.

### *Statistics*

Non-parametric correlations were used (Spearman's) because headache scores were not normally distributed.  $\chi^2$  tests were used to look for associations between cavernous sinus invasion and the presence or absence of headache. Statistical significance was assessed at the  $P < 0.05$  level. Statistical analyses were computed on commercial software: SPSS version 9.0.

## **2.1.4 Results**

### *Headache*

The prevalence of headache was 70% (44/63) and the raw data are shown in Table 12. The clinic headache scores closely matched the scores obtained from the prospective headache diaries (Figure 9;  $r = 0.93$ ,  $P < 0.005$ ) thus validating the semi-quantitative assessment of headache. The headache scores within each tumour sub-type are shown in Table 13. There was a significant association between pituitary tumour-associated headache and the presence of a family history of headache ( $\chi^2 = 8.36$ ,  $P = 0.004$ , Table 14).

### *Pituitary volume*

There was no positive correlation between pituitary volume and headache ( $r = -0.32$ ,  $P = 0.01$ ; Figure 6).

### *Cavernous Sinus Invasion*

There was no association between headache and any compartment of cavernous sinus invasion (Table 14).

**Table 12. Headache score, tumour volume and cavernous sinus invasion**

Patient code	Tumour type	Headache		Tumour Volume (mls)	C.S Invasion		
		Y/N	Score		Lateral	Inferior	Superior
1	NFA	1	240	5.1	1	1	1
2	Cushings	1	32	0.4	0	0	0
3	NFA	1	384	1.3	0	0	0
4	Cranio	1	344	17.8	0	0	0
5	Cranio	1	672	15.1	0	0	0
6	NFA	0	0	3.1	0	0	0
7	Prl	1	1536	0.2	0	0	0
8	NFA	1	224	1.2	1	1	1
9	Prl	1	240	0.9	0	1	0
10	Prl	1	390	2.1	0	0	0
11	Cushings	1	64	0.1	0	0	0
12	NFA	0	0	0.6	0	0	0
13	Acro	0	0	0.5	0	0	0
14	Prl	1	2021	0.2	0	0	0
15	NFA	1	24	6.6	0	1	1
16	Acro	0	0	1.2	0	0	0
17	Acro	0	0	10.2	1	0	1
18	Cranio	1	880	2.4	0	0	0
19	Prl	1	75	4.2	1	1	1
20	NFA	0	0	13.2	0	0	0
21	Prl	1	1728	0.3	0	0	0
22	Prl	1	720	6.5	1	1	1
23	Prl	1	20	3.2	0	0	0
24	Acro	1	194	7.0	0	1	0
25	Cushings	0	0	1.0	0	0	0
26	NFA	0	0	14.9	1	0	1
27	NFA	1	520	3.3	1	1	1
28	Prl	1	305	0.07	0	0	0
29	NFA	0	0	3.1	0	0	0
30	NFA	0	0	10.3	0	0	0
31	Prl	1	384	0.1	0	0	0
32	Acro	0	0	0.65	0	0	0
33	NFA	0	0	10.7	1	1	1
34	Acro	0	0	3.9	0	0	0
35	TSH	1	1800	1.7	1	0	1
36	NFA	1	80	1.0	0	0	0
37	TSH	0	0	0.5	0	0	0
38	Prl	1	166	0.4	0	0	0
39	Acro	1	2160	0.9	0	1	0
40	Acro	1	3320	2.6	0	0	0
41	NFA	1	400	1.2	0	0	0
42	Cushings	1	240	0.1	0	0	0
43	Cushings	1	360	0.2	0	0	0
44	NFA	1	90	3.2	1	1	1
45	Prl	1	192	2.7	0	0	0
46	Prl	1	848	0.1	0	0	0
47	Prl	1	1476	0.1	0	0	0
48	Prl	1	656	0.1	0	0	0
49	NFA	0	0	5.1	0	1	0
50	Prl	1	2	9.4	1	1	1
51	Acro	1	180	4.6	0	0	0
52	Prl	1	1440	0.1	0	0	0
53	NFA	1	1104	8.7	1	0	1
54	Acro	1	360	0.1	0	0	0
55	Acro	0	0	0.23	0	0	0
56	NFA	1	264	9.0	0	0	0
57	Acro	1	2880	0.1	1	1	0
58	Prl	1	60	1.5	0	0	0
59	Cranio	1	216	0.7	0	0	0
60	Cranio	0	0	3.6	0	0	0
61	Acro	0	0	0.3	1	0	1
62	TSH	0	0	7.7	0	0	0
63	NFA	1	160	6.5	0	0	0

NFA non-functioning adenoma, Prl prolactinoma, Acro acromegaly, Cranio craniopharyngioma, TSH TSHoma

**Table 13. Headache scores for each tumour sub-type**

<b>Tumour sub-type</b>	<b>n</b>	<b>Mean score</b>
Growth hormone secreting tumour	12	758
Prolactinoma	17	659
TSHoma	3	600
Craniopharyngioma	5	362
Non functioning adenoma	20	194
ACTH secreting tumour	6	200

**Table 14. Association between cavernous sinus invasion, family headache history and headache**

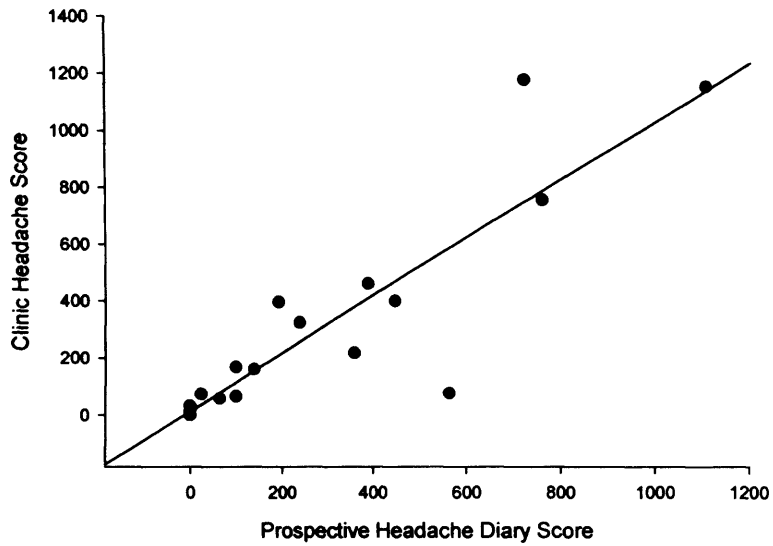
<b>Cavernous Sinus Invasion</b>	<b><math>\chi^2</math> test</b>	<b>Significance</b>
Superior	0.17	NS
Inferior	0.17	NS
Lateral	1.08	NS
<b>Family History</b>	<b>8.36</b>	<b><math>P = 0.004^*</math></b>

NS  $P > 0.05$

\* statistically significant association

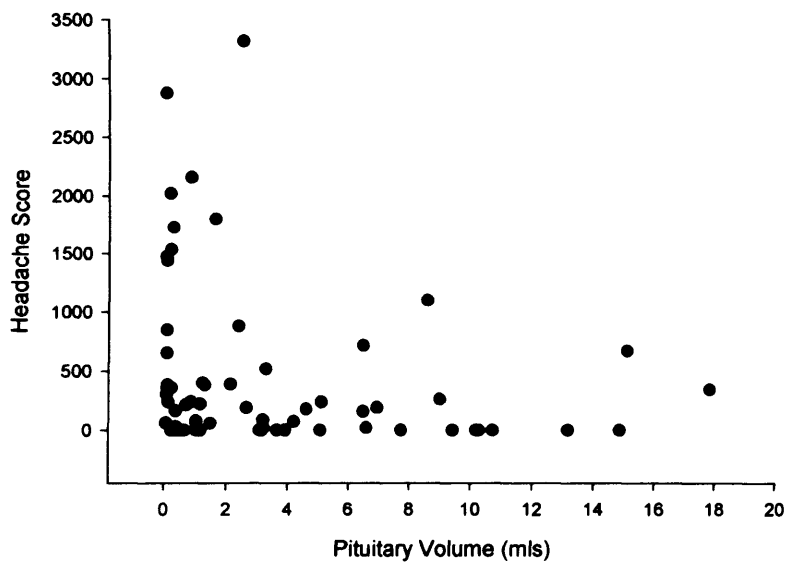
**Figure 9. Validation of retrospective headache score**

**Clinic Headache Score vs Prospective Headache Diary Score**



**Figure 10. Relationship between pituitary volume and headache**

**Headache Score against Pituitary Volume**



### 2.1.5 Discussion

The close correlation between the retrospective clinic headache scores and the prospective headache diaries validates the semi-quantitative assessment of headache in these patients. The clinic headache score is a modification of a well-recognised assessment of headache morbidity (Stewart et al., 2000) and the high scores confirms the clinical impression that patients with pituitary tumours have a significant headache problem.

No positive correlation between headache and tumour volume was found, which would have been expected if the stretching of adjacent dural structures, such as the wall of the cavernous sinus or diaphragma sellae were an important mechanism of pituitary-associated headache. Headache also did not correlate with cavernous sinus invasion or carotid encasement. There was in particular no association between headache and invasion of the lateral compartment of the cavernous sinus ( $\chi^2 = 1.08$ ), which contains the ophthalmic and maxillary divisions of the trigeminal nerve. The lack of association between cavernous sinus invasion and headache is in agreement with a previous study (Abe et al., 1998). Headache was also unrelated to the degree of carotid encasement, a potential indicator of tumour interference with the sympathetic plexus surrounding the internal carotid.

The 70% prevalence of headache in patients with pituitary tumours is significantly higher than the prevalence of headache in the general population (Rasmussen and Olesen, 1992; Lipton et al., 2002) confirming that there is a pro-nociceptive quality attached to pituitary tumours. Mean headache scores were high in the acromegaly and prolactinoma group, although larger numbers are required for a true comparison between tumour sub-types.

The significant association between a family headache history and pituitary-associated headache suggests that genetic factors are important in predicting whether a patient with a pituitary tumour will develop headache as part of the presentation. As discussed in Section 1.4, it is known that first-degree relatives of migraine sufferers are also predisposed to migraine (Gervil et al., 2001). The presence of a pituitary



tumour may simply be the trigger in a patient who is already pre-disposed to headache.

Clearly functional imaging and animal studies suggest that the hypothalamo-pituitary axis plays an important role in the pathophysiology of primary headache syndromes such as migraine and the TACs, particularly cluster headache. As outlined in Section 1.4, in migraine there is evidence of hypothalamic dysfunction (Peres et al., 2001) and the presence of polyphagia, polyuria and polydipsia as part of the premonitory phase of migraine further implicates the hypothalamus (Giffin et al., 2003). The striking relationship between the menstrual cycle and migraine in women suggests that changes in the hypothalamo-pituitary axis can profoundly alter the expression of migraine (Silberstein, 2000a). That functional alterations in the hypothalamus may be relevant to headache has also been reviewed earlier in reference to cluster headache (May et al., 1998a; Goadsby and May, 1999) and SUNCT (May et al., 1999b) where there is evidence of hypothalamic activation during a bout (Section 1.5). The elucidation of the trigemino-hypothalamic tract (Malick et al., 2000) provides anatomical evidence for a direct neuronal interaction between the hypothalamo-pituitary axis and the trigeminal nucleus.

Although there was no significant association between cavernous sinus invasion and headache for the whole group, headache was ipsilateral to the side of cavernous sinus invasion in four cases. This appears to be in keeping with the clinical observation that there may be sub-set of patients with pituitary disease who have headache ipsilateral to the side of cavernous invasion (de Groot & Jameson, 2000). If this clinical impression is correct, it is unclear why cavernous sinus invasion should only cause headache in a select proportion of patients, and may suggest that headache associated with cavernous sinus invasion requires other factors in addition in order to give rise to headache.

This was an improvement on the previous study (Abe et al., 1998) because tumour volume rather than tumour diameter was measured, cavernous sinus invasion was classified by previously validated criteria, and a quantitative estimate of headache was calculated and correlated with tumour size. However, although there was a close statistical correlation between the prospective diary scores and the retrospective

headache scores, the quantification of headache is likely to have produced the most inaccuracies, as the latter necessarily has a subjective component on behalf of the patient. A further possible confounding variable in this study arises from the potentially incorrect assumption that all patients experienced headache directly as a result of pituitary disease. Although all patients reported headache onset coincident with other classical pituitary tumour symptoms, it is possible that there were undetected aetiological causes for headache in certain cases. A larger series is required to allow for these potential confounders.

In summary, I found no association between pituitary size and headache, nor was there an association between cavernous sinus invasion and headache. This study may be important in dispelling the currently accepted view that headache in pituitary tumours is primarily a structural problem. Further studies are required to determine the pathophysiology of pituitary tumour-associated headache, such as the importance of biochemical properties of the tumour, which is now investigated.

### **3. Pituitary tumours and headache: biochemical factors**

### **3.1 Study 2: the association between calcitonin gene related peptide, substance P and headache in pituitary tumours**

Levy MJ, Classey J, Maneesi S, Powell M, Meeran K, Goadsby PJ

Abstract ref: Cephalalgia 2003; 23: 687

#### **3.1.1 Abstract**

*Objectives:* To determine if the differential expression of Calcitonin Gene Related Peptide (CGRP) or substance P (SP) in a range of pituitary tumours was related to the presence or absence of headache.

*Methods:* Using recognised immunohistochemical techniques, twenty-six consecutive pituitary adenoma specimens were examined for the presence of CGRP and SP. One normal post mortem pituitary specimen was included for comparison. A separate observer divided the patients into two groups: headache and non-headache. The association between the presence of CGRP, SP and headache was observed.

*Results:* CGRP was observed in seven specimens (27%) and SP in six tumour specimens (23%), with cytoplasmic staining being the predominant morphological picture. CGRP and SP were co-expressed in the same tumour specimen in five cases. There was no significant association between the presence of CGRP and headache ( $\chi^2$  0.86;  $P = 0.35$ ). CGRP and SP were not observed in the control specimen. There was no correlation between tumour subtype and the presence of CGRP or SP.

*Conclusions:* The mechanism of pituitary tumour-associated headache remains undetermined. The significance of the presence of CGRP and SP in pituitary tumours is unknown but does not appear to be related to headache or endocrine activity of the tumour.

### **3.1.2 Introduction**

Study 1 demonstrated that the physical properties of pituitary tumours are not associated with headache in the majority of patients. Given the involvement of neuropeptides in primary headache (Edvinsson and Goadsby, 1995), and the observation that headache appears to be particularly prevalent in acromegaly (Ezzat et al., 1994; Couch, 1986) and prolactinoma (Abe et al., 1998), a biochemical explanation for headache is possible. The previous suggestion by several groups that pituitary tumour-associated headache may be due to the production of a pro-nociceptive peptide (Williams et al., 1986; Pascual, 2000) has been partly driven by the observation that somatostatin analogues can have an immediate and dramatic analgesic effect on the headache associated with acromegaly (Pascual et al., 1991; Newman et al., 1998) and prolactinoma (Williams et al., 1987). Moreover, the uncoupling of the growth hormone from headache in acromegaly (Webb et al., 1989; Levy et al., 2003b) suggests that biochemical factors other than growth hormone may be involved in the headache associated with certain pituitary tumours.

As has been discussed in the general introduction, advances in the understanding of primary headache pathophysiology have pointed towards certain peptides as being important in the transmission of cranial nociceptive information (Goadsby and Edvinsson, 1993). Calcitonin gene-related peptide (CGRP), which rises during migraine (Goadsby et al., 1990; Gallai et al., 1995) and cluster headache (Goadsby and Edvinsson, 1994; Fanciullacci et al., 2000), could theoretically give rise to headache in pituitary tumours by acting on CGRP-containing sensory fibres present within the cavernous sinuses (Keller and Marfurt, 1991) and cerebral arteries (Uddman et al., 1985). Although substance P (SP) levels do not rise during migraine (Goadsby et al., 1990; Gallai et al., 1995) or cluster headache (Goadsby and Edvinsson, 1994; Fanciullacci et al., 2000), SP-immunoreactive fibres richly innervate the dura mater in the cavernous sinus (Keller and Marfurt, 1991) and are present in the dorsal horn of the trigeminal nucleus (Ribeiro-da-Silva and Hokfelt, 2000) suggesting it may have a role in headache. Sumatriptan, an agent that is useful in migraine and cluster headache (Ferrari et al., 2001), is also effective in the headache associated with pituitary tumours, possibly by the inhibition of vasoactive peptides, such as CGRP and SP, which may be theoretically produced by the tumour

(Pascual, 2000). The review in Section 1.10 has presented the evidence that CGRP (Steel et al., 1992) and SP (Jessop et al., 1992) have paracrine roles within the hypothalamo-pituitary axis, and have been isolated in tumorous and non-tumorous human pituitary tissue (Roth and Krause, 1990; Wimalawansa, 1994). In the current study, I examined the expression of CGRP and SP within pituitary tumours, hypothesising that it may predict headache.

### **3.1.3 Subjects and Methods**

#### *Patient demographics*

Twenty-six consecutive patients with pituitary tumours presenting to the single dedicated pituitary neuro-surgical unit, in which this thesis was based (QS), were included in the study (18 female, 9 male; mean age  $51.9 \pm 2.9$ ). The diagnoses of the patients entering the study included acromegaly (n = 7), Cushing's disease (n = 4), prolactinoma (n = 2) and non-functioning adenoma (n = 13).

#### *Radiological characteristics*

All patients had pre-operative MRI scans that showed evidence of pituitary lesions. Macro-adenomas were present in 20 cases, the remainder having micro-adenomas (n = 6). Cavernous sinus invasion was present in 6 tumours.

#### *Study protocol*

All patients were interviewed pre-operatively and the presence or absence of headache was documented. A headache history was obtained and a phenotypic description was given according to International Headache Society (I.H.S) diagnostic criteria (2004). Pituitary tissue was obtained at surgery and immediately fixed in 4% paraformaldehyde and stored at 4 ° C. Local ethical committee approval of the protocol was obtained, and tissue was anonymised for immunohistochemical study. One post mortem pituitary specimen was available as control tissue (kindly provided by Professor F. Scarivelli, Institute of Neurology).

#### *Immunohistochemistry*

##### **Tissue collection and preparation**

Pituitary tissue was obtained at operation from consecutive patients undergoing transphenoidal surgery. Tumour specimens were kindly provided by the lead pituitary surgeon in the hospital (Mr M Powell) and the tissue was given an anonymous code. Perioperatively, the specimens were immediately transferred into 4% paraformaldehyde fixative and refrigerated for 24 hours at 4°C. The tissue was subsequently transferred to ½ paraformaldehyde/ ½ 30% sucrose for 48 hours and then stored in 30% sucrose alone at 4°C prior to immunostaining.

### *Immunostaining method*

#### 1. Xylene Removal of Paraffin and rehydration

After collection and storage, the pituitary tissue was paraffin-embedded, and sections were cut and slide-mounted (6 µm). Paraffin was removed from the sections by the addition of xylene followed by rehydration. The selected slides, containing 2 sections of pituitary tissue, were placed in plastic racks with handles through a series of 2 x xylene, and subsequently treated with successively lower-graded alcohols (2 x 100, 2 x 95%, 1 x 80% and 1 x 50%). Slides were then placed in tap water and rinsed to remove the alcohol. This was followed by immersion in distilled water and then 3 x 5 minute rinses in 0.01 M phosphate-buffered saline (PBS).

#### 2. Removal of Endogenous Peroxidases

Following paraffin removal and rehydration, endogenous peroxidases were removed by oxidation. Slides were incubated in 0.6% hydrogen peroxide for 30 minutes and washed in PBS for 3 x 5 minutes.

#### 3. Antigen Retrieval

Heat treated antigen retrieval was performed to remove paraffin bonds and increase the sensitivity of the immunoreaction (Cattoretti et al., 1993). Slides were placed in covered containers filled with citrate buffer (pH =7.4) and heated in the microwave for 10 minutes at full power, in two 5 min bouts. Further buffer was added after the first heating bout, in order to avoid dehydration of the sections. Sections were then cooled safely through heat exchange, by running cold water into the containers without exposing the slides to the air.

#### 4. Formation of water-resistant barrier

The slide-bound sections were re-washed for 3 x 5 minutes in PBS and placed into an incubation-box, whose absorbent base had been soaked with distilled water. Both sections on each slide were circled with a peroxidase-anti-peroxidase pen to prevent the reagents from spreading over the slide and to avoid unnecessary wastage of reagents.

#### 5. Addition of blocking agent

Sections were incubated with a blocking-agent in order to increase the specificity of the immunoreaction (Polak and Van Noorden, 1987). Blocking agent involves the use of phosphate buffered serum derived from a species different from that in which the primary antibody was raised, to avoid non-specific binding to the primary antibody. In both the CGRP and SP reactions, phosphate-buffered goat serum, derived from diluting normal goat serum in phosphate buffered saline, was used as the blocking agent as both primary antibodies were raised in rabbit. Blocking agent was made up at a dilution of 1:20 (1 ml of reconstituted frozen serum was added to 19 ml of phosphate buffered saline) and added to each section (1 ml). A box containing the slides was covered with its lid and left at room temperature for 1 hour.

#### 6. Removal of blocking agent

Blocking agent was tipped off into a waste-collection beaker for separate disposal, as well as any excess of the sodium azide-containing normal serum.

#### 7. Addition of Primary Antibody

Because I was interested in the level of CGRP and SP peptide secretion, the presence of peptide rather than receptor was examined. Primary antibody was made-up at the appropriate dilution in phosphate buffered serum. Several dilutions were attempted for each antibody until the optimum dilution was found to compromise between background and specific staining. Anti-rat CGRP polyclonal antibody raised in rabbit (Amersham Biosciences, UK) was diluted to an optimal dilution of 1:1000. Antihuman SP polyclonal antibody raised in rabbit (Biogenesis Ltd, UK) was diluted to an optimal dilution of 1:400. Primary antibody (200 µl) was added to each slide



and the incubation box covered with its lid and incubated overnight in the refrigerator at 4°C.

#### 8. Addition of Secondary Antibody

Biotinylated secondary antibody was made up to a dilution of 1:30, i.e 10 µl of secondary antibody in 2 ml of phosphate buffered serum and added to the slide. The sections were left to incubate for 1 hour in a covered box at room temperature.

#### 9. Addition of Tertiary Antibody

Secondary antibody was washed off with 3 x 5 minute rinses with phosphate buffered saline. The avidin-biotin complex (ABC) as the tertiary antibody was used (Boenish, 1989). In this method, ABC is added to the secondary antibody, where open avidin sites bind to the biotin component of the secondary antibody. Because a large number of biotin molecules can be attached to a single antibody, this method amplifies the signal of the antigen-primary antibody-secondary antibody complex, allowing for increased sensitivity of the immunostain (Polak and Van Noorden, 1987). The ABC reagent was constituted 30 minutes prior to addition to the section, in which 2 drops of solution A were added to 2 drops of solution B from the Standard Elite Vector ABC kit, in 5 ml phosphate buffered saline. The ABC reagent was added each slide (200µl), and the sections covered with a lid and left to incubate at room temperature for 1 hour. After the reaction, the ABC solution was washed off with 3 x 5 minute rinses of phosphate buffered saline.

#### 10. Diaminobenzidine Reaction

The diaminobenzidine (DAB) reaction causes colour change and allows the antigen-antibody complex to be visualised. In this reaction, hydrogen peroxide is used as the substrate and DAB as the chromogen (Polak and Van Noorden, 1987). The oxidised DAB polymerises and forms an insoluble dark brown precipitate at the site of the reaction. In certain situations, the brown colouration may not be sufficiently visualised, in which case nickel ( $\text{Ni}^{2+}$ ) may be added to the incubation medium to produce a black product (Polak and Van Noorden, 1987).

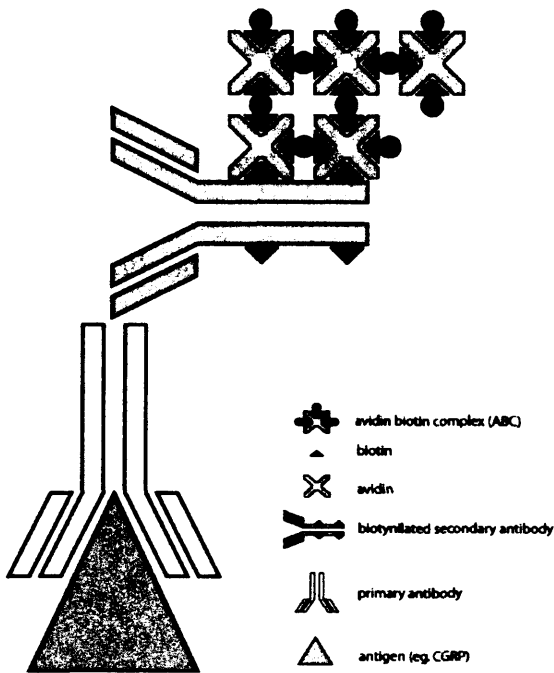
$\text{Ni}^{2+}$ -enhanced or non-  $\text{Ni}^{2+}$ -enhanced half-strength DAB solution was made up immediately prior to adding to the sections, using a standard Vector DAB kit. This consisted of 5 ml of distilled  $\text{H}_2\text{O}$  + 2 drops of buffer stock solution (mix) + 2 drops of DAB stock solution (mix) + 1 drop of  $\text{H}_2\text{O}_2$  (mix) + 1 drop of  $\text{Ni}^{2+}$  solution (mix). For the SP reaction, chromagen nickel-enhanced half-strength DAB solution was used, and for CGRP, the DAB reaction was not nickel-enhanced. The reaction was timed after the addition of DAB solution (200  $\mu\text{l}$ ) to each slide until an appropriate level of differentiation was seen, as black / grey reaction product in  $\text{Ni}^{2+}$ -enhanced chromagen and brown in non-  $\text{Ni}^{2+}$ -enhanced chromagen. The reaction was aborted by placing the sections in deionised  $\text{H}_2\text{O}$ . On average, sections were reacted with DAB for between 5-10 minutes. A schematic diagram of the avidin-biotin complex binding to the primary and secondary antibody complex is shown in Figure 11.

#### 11. Rehydration and Cover Slipping of Sections

The sections were dehydrated by placing them through increasing concentrations of alcohol (50%, 80%, 2 x 95%, 2 x 100%) and then 2 x xylene (the reverse of step 1). The sections were finally cover slipped using DPX mountant and left to dry before inspection.

Control sections were prepared using the same method as above with the omission of primary antibody. All control sections showed no background staining. The specificity of CGRP and SP anti-bodies was confirmed using human spinal cord dorsal horn as a positive control (Figure 12).

**Figure 11. Schematic diagram of avidin-biotin complex**



### *Antibody specificity*

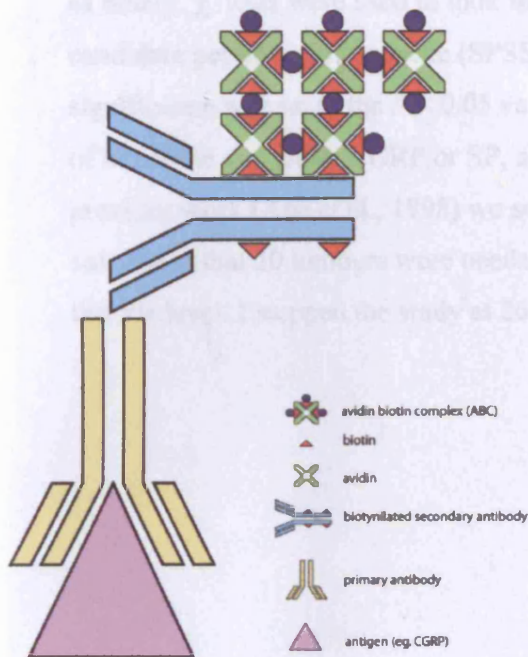
In accordance with Burry (2000), antibody specificity was confirmed by replacing the primary antibody with normal goat serum, and using positive control tissue. Human post-mortem spinal cord, which is known to contain CGRP and SP, was used as the positive control, and was reacted with primary antibody. No background staining was observed in any specimens in the absence of primary antibody, and specificity for CGRP and SP was confirmed by the observation of axonal and dendritic staining in human spinal cord (Figure 12).

### *Identification of CGRP / SP positive cells*

Analysis of the tissue was performed by two independent observers who were blinded to headache status (the candidate and Dr S Manesri) to exclude inter-observer variation. The presence or absence of immuno-positivity was documented for each specimen and a comment made about the morphology and distribution of the staining. After the data was locked, the association between the presence of peptide and the presence of headache was studied.

### *Statistics*

**Figure 11. Schematic diagram of avidin-biotin complex**



### *Antibody specificity*

In accordance with Burry (2000), antibody specificity was confirmed by replacing the primary antibody with normal goat serum, and using positive control tissue. Human post-mortem spinal cord, which is known to contain CGRP and SP, was used as the positive control, and was reacted with primary antibody. No background staining was observed in any specimens in the absence of primary antibody, and specificity for CGRP and SP was confirmed by the observation of axonal and dendritic staining in human spinal cord (Figure 12).

### *Identification of CGRP / SP positive cells*

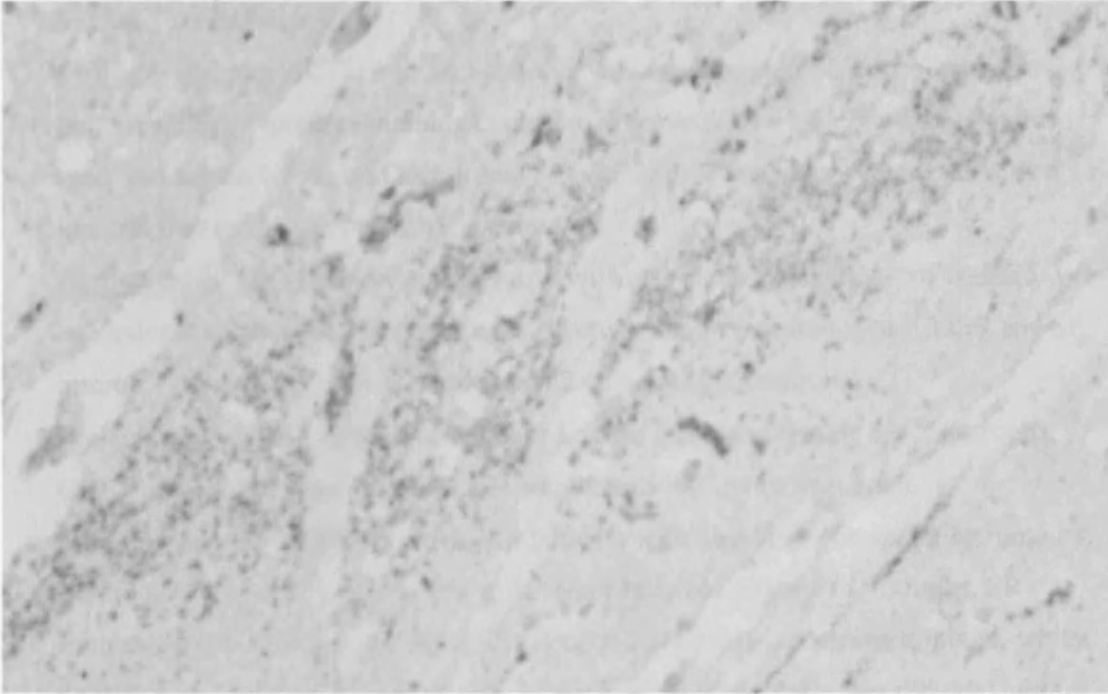
Analysis of the tissue was performed by two independent observers who were blinded to headache status (the candidate and Dr S Manesri) to exclude inter-observer variation. The presence or absence of immuno-positivity was documented for each specimen and a comment made about the morphology and distribution of the staining. After the data was locked, the association between the presence of peptide and the presence of headache was studied.

### *Statistics*

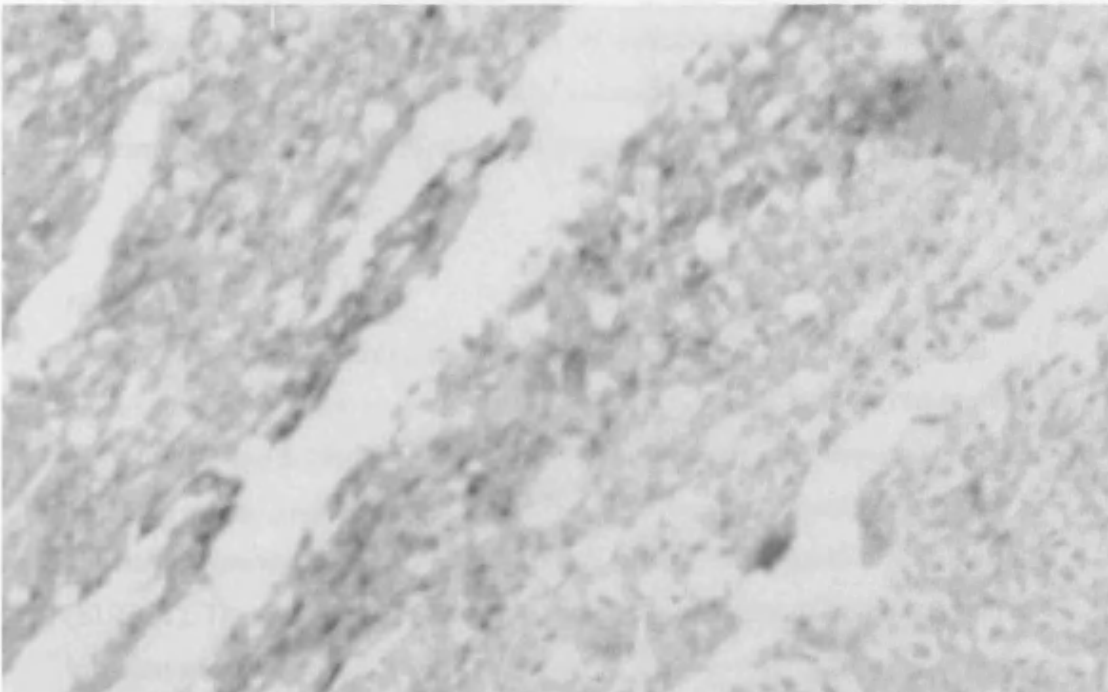
The presence of headache and CGRP/ SP immunopositivity were recorded and treated as binary.  $\chi^2$  tests were used to look for an association between the presence of candidate peptide and headache (SPSS Version 10 Chicago IL). Statistical significance was set to the  $P < 0.05$  value. Assuming a greater than chance association of headache and either CGRP or SP, and a headache frequency of 50% based on previous work (Abe et al., 1998) we sought a 30% or greater association. It was calculated that 20 tumours were needed for an 80% power of detecting a difference at the 5% level. I stopped the study at 26 tumours.

**Figure 12. Specificity of CGRP and substance P antibody in dorsal horn of human spinal cord**

3.1.4 Results



**Figure 12a. CGRP immuno-staining in dorsal column of human spinal cord**



**Figure 12b. SP immuno-staining in dorsal column of human spinal cord**

### 3.1.4 Results

#### *Immunohistochemistry*

Cushings disease: ACTH immuno-staining was confirmed in all 4 specimens. 3 tumours elicited immuno-staining exclusively for ACTH, whilst one also contained scattered somatotrophs and lactotrophs. CGRP and SP immuno-staining cells were identified in one ACTH-positive specimen.

Acromegaly: GH immuno-staining was confirmed in all 7 specimens, of which 5 were exclusively somatotroph tumours, and 2 were somato-mammotrophs. CGRP and SP immuno-staining cells were identified in 2 somatotroph tumours.

Prolactinomas: Both specimens displayed dense immuno-staining for lactotrophs. CGRP and SP immuno-staining cells were identified in one specimen.

Non-functioning adenomas: Of the 13 patients with non-hormone secreting tumours, 11 did not stain for any hormones and 2 were found to contain thyrotrophs. SP immuno-reactive cells were identified in both thyrotroph-containing tumours, whilst CGRP was identified in one of these. In the remaining tumours that did not stain for any hormones, CGRP immuno-reactivity was identified in 2 specimens.

CGRP-containing cells were characterised morphologically by predominantly cytoplasmic staining (Figure 13), and the SP-containing cells appeared as a combination of nuclear and cytoplasmic staining (Figure 14). In both cases, the immuno-positive cells were distributed evenly throughout the tumour tissue.

#### *Headache*

For the whole group, headache was present in 11 patients (42%). Headache prevalence was highest in prolactinoma (100%) and acromegaly (43%) but was seen in all tumour types (Table 1). The commonest headache phenotype was migraine (n=10). One patient presented with an I.H.S diagnosis of Short lasting headache Unilateral Neuralgiform headache attacks Conjunctival congestion and Tearing (SUNCT). There was no association between the presence of cavernous sinus invasion and headache.

*Relationship between CGRP, SP and headache*

There was no significant association between the presence of CGRP and headache ( $\chi^2 = 0.86$   $P = 0.35$ ), nor was there an association between the presence of SP and headache ( $\chi^2 = 0.19$ ;  $P = 0.67$ ).



**Table 15. Tumour, headache and immuno-staining characteristics**

IMMUNO	SEX	TUMOUR SIZE	INVASION	HEADACHE	PHENOTYPE	CGRP	SP
ACTH	F	MACRO	-	-	-	+	+
GH	F	MICRO	-	-	-	-	-
NFA	F	MACRO	+	+	MIGRAINE	-	-
NFA	F	MACRO	+	+	MIGRAINE	-	-
NFA	M	MACRO	-	-	-	-	-
TSH	F	MACRO	-	+	MIGRAINE	+	+
NFA	M	MACRO	-	-	-	-	-
GH/PRL	M	MICRO	-	-	-	-	-
NFA	F	MACRO	-	-	-	-	-
GH	M	MICRO	-	+	MIGRAINE	+	+
PRL	F	MACRO	-	+	MIGRAINE	-	-
NFA	F	MACRO	-	-	-	+	-
NFA	F	MACRO	-	-	-	-	-
NFA	F	MACRO	-	-	-	-	-
NFA	M	MACRO	+	-	-	-	-
PRL	M	MACRO	-	+	MIGRAINE	+	+
GH	F	MICRO	+	-	-	+	+
NFA	F	MACRO	-	+	MIGRAINE	+	-
ACTH	F	MACRO	-	+	MIGRAINE	-	-
GH/PRL	F	MACRO	+	+	MIGRAINE	-	-
ACTH	M	MICRO	-	-	-	-	-
GH	F	MACRO	-	+	MIGRAINE	-	-
ACTH	F	MICRO	-	-	-	-	-
TSH	F	MACRO	+	-	-	-	+
NFA	M	MACRO	-	+	SUNCT	-	-
GH	F	MACRO	-	-	-	-	-

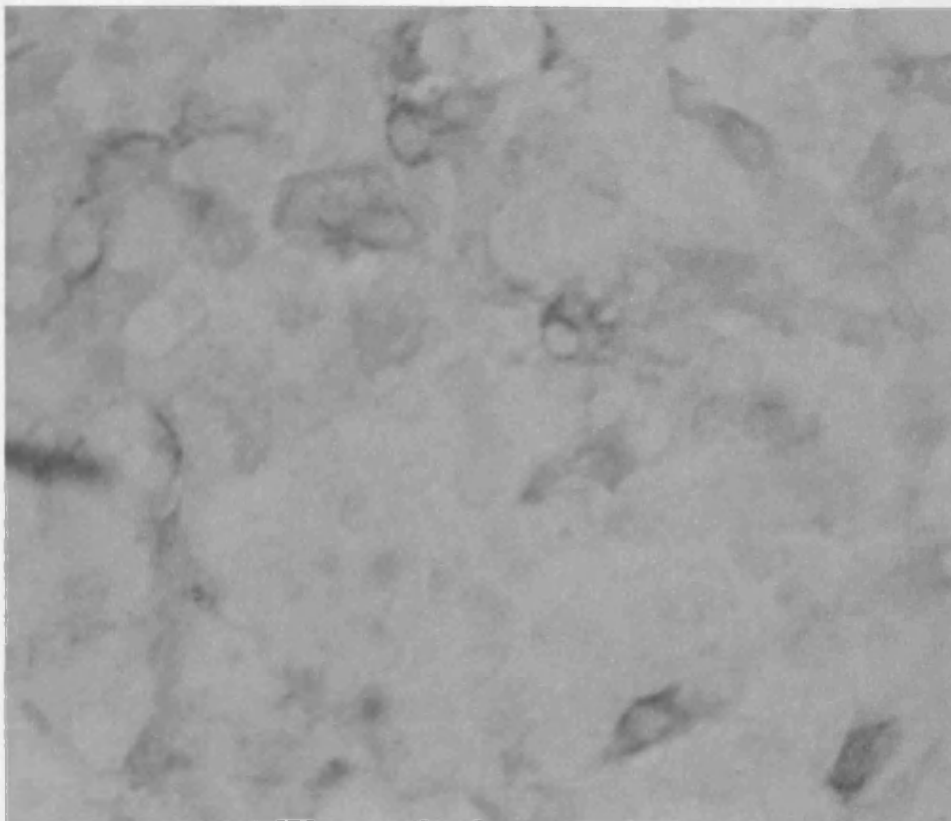
NFA= non functioning adenoma, GH = growth hormone secreting tumour, PRL = prolactinoma, GH/ PRL = mixed growth hormone/ prolactin secreting (somatomammotroph), TSH = thyrotroph containing, SUNCT = Short lasting Unilateral Neuralgiform attacks with Conjunctival congestion and Tearing.

**Table 16. Headache and immunopositivty within each tumour sub-type**

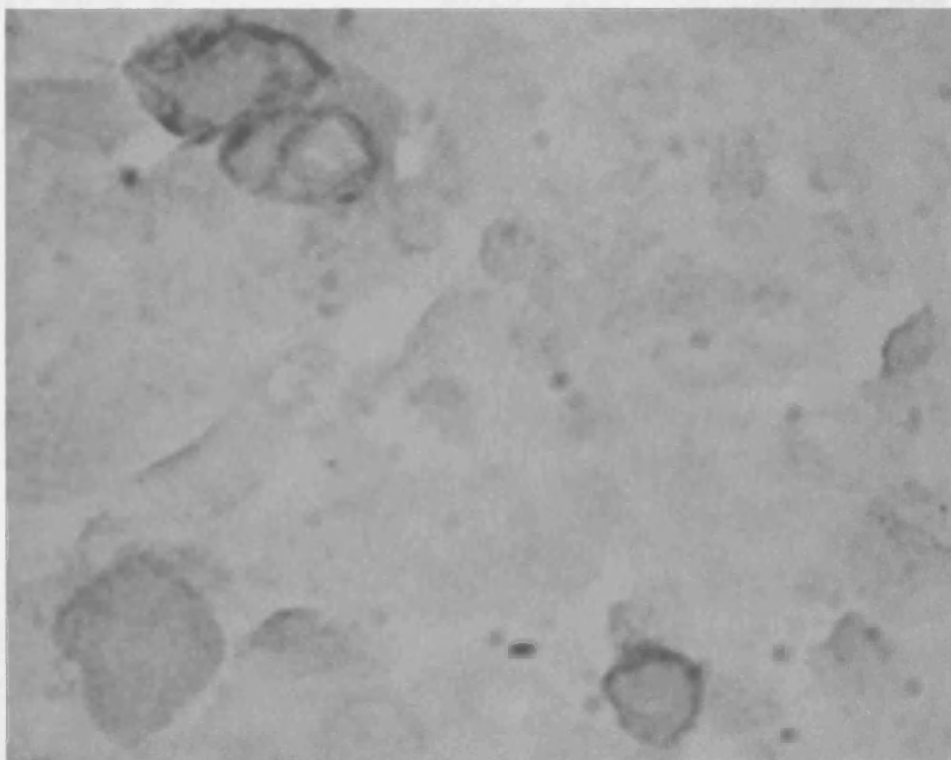
Tumour sub-type	Total no.	Headache +ve	CGRP +ve	SP +ve
ACTH	4	1	1	1
GH	5	2	2	2
GH/PRL	2	1	0	0
PRL	2	2	1	1
TSH	2	1	1	2
NFA	11	4	2	0
Total	26	11	7	6

NFA= non functioning adenoma, GH = growth hormone secreting tumour, PRL = prolactinoma, GH/ PRL = mixed growth hormone/ prolactin secreting (somatomammotroph)

**Figure 13. CGRP immuno-staining appearances**

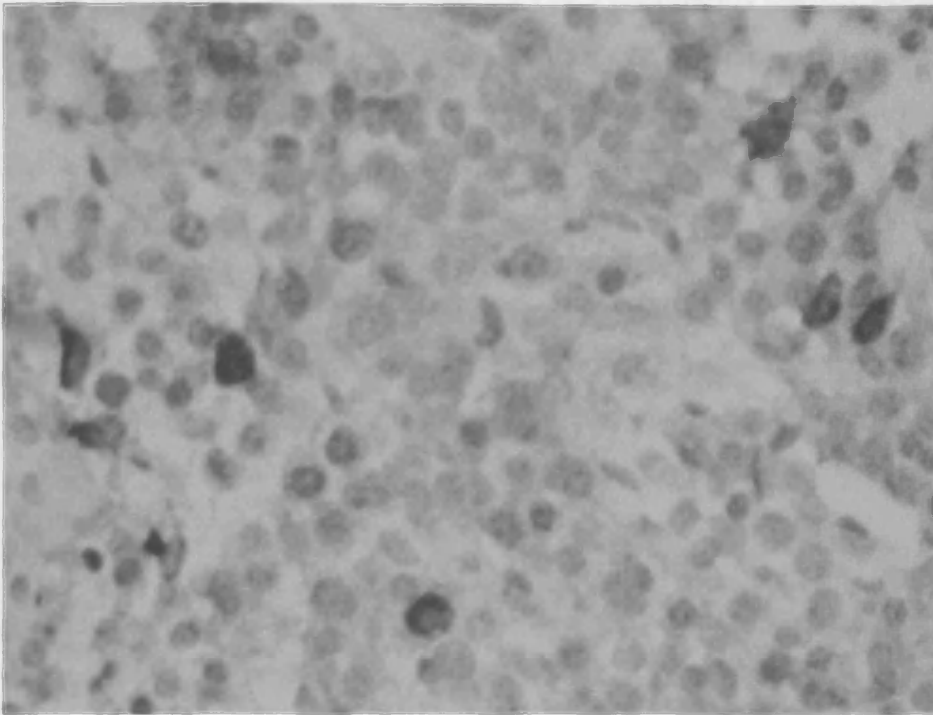


**Figure 13a Non-functioning adenoma (x20)**

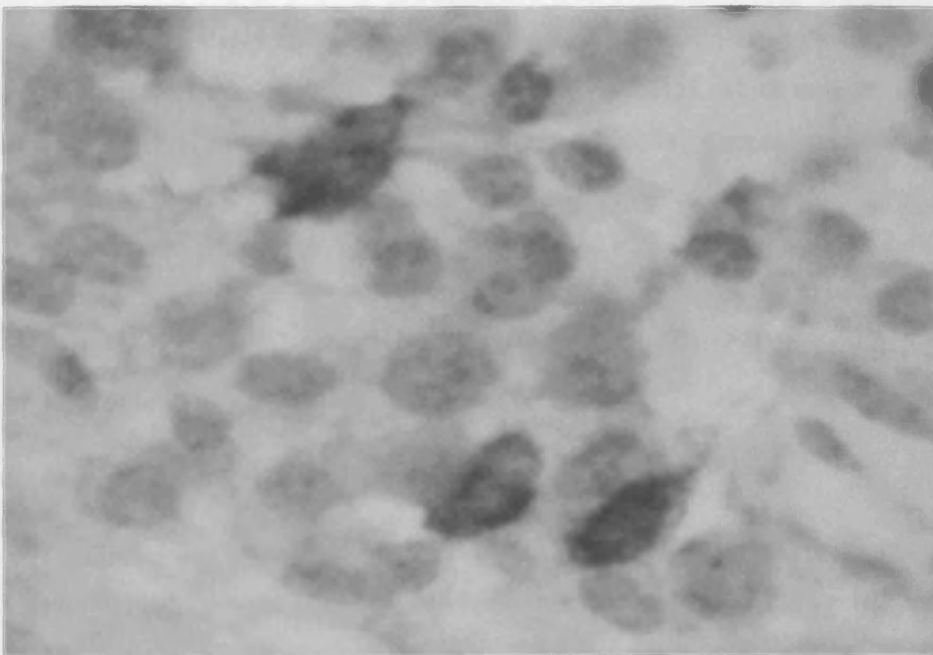


**Figure 13b. Non-functioning adenoma (x40)**

**Figure 14. Substance P immuno-staining appearances**



**Figure 14a. Non-functioning adenoma (x20)**



**Figure 14b. Non-functioning adenoma (x40)**

### **3.1.5 Discussion**

The results of this study do not demonstrate a relationship between the presence of CGRP or SP with headache. Although the tumour numbers in this study were relatively small, the study was powered for an association that would be robust, so the findings show that headache can be present without any demonstration of CGRP or SP and *vice versa*. I did not find any association between CGRP or SP with tumour activity, although the presence of SP in association with thyrotrophs has been reported by other groups (Roth et al., 1994; Arita et al., 1994). The co-localisation of CGRP and SP in 5 out of 9 immuno-positive tumours is in keeping with previous observations that CGRP and SP have a tendency to co-localise (Ribeiro-da-Silva et al., 2000), although this has not been previously described in relation to the anterior pituitary gland.

The aim of the study was to determine whether the presence of CGRP and SP could predict pituitary tumour-associated headache. The result was negative and this must raise study design issues. An underlying assumption of this, and similar studies that seek associations with headache, is that the headache is a direct result of the pituitary tumour. It is possible other variables impinged on that likelihood to manifest headache. For example, a genetic predisposition to primary headache and the presence of environmental trigger factors was not documented, which may have been important in individual cases (Goadsby et al., 2002). Migraine is more commonly seen in women (Goadsby et al., 2002), and the fact that the ratio of women to men in the study group was greater than 2:1 may have biased the results. Also, I did not take into consideration the possibility that endocrine deficiency could have been a confounding variable, although no patients exhibited typical features of cortisol deficiency headache.

Headache was simplistically treated as a binary variable (present or absent), whereas a clinical sub-categorisation of headache phenotypes may have been more meaningful. This would require a much larger study to attempt to associate CGRP or SP with a particular type of headache. Although headache classification was not the primary focus of the study, being fully investigated in Chapter 4 of this thesis, migraine was

the commonest headache phenotype observed in this study. The other case presenting with headache had a diagnosis of SUNCT. If pituitary tumour-associated headache is a heterogeneous rather than a homogeneous clinical entity, much higher numbers would be required to comment on the relative prevalence of each phenotype within particular tumour sub-groups.

The presence of peptide rather than receptor status within the tumour tissue was studied because the hypothesis generated was that pituitary tumour-associated headache is caused by active peptide secretion. Although the importance of immunoblotting as an antibody purity determinant is acknowledged, antibody specificity was demonstrated by the use of both a negative and positive control (Burry, 2000). However, molecular techniques such as *in situ* hybridisation may have provided more sensitivity to this study. It is possible that the immunohistochemical method used was insufficiently sensitive to demonstrate small quantities of CGRP and SP that may have been present in some of the apparently negative tumour specimens. Further studies looking at mRNA rather than presence of peptide may be necessary if the nociceptive peptide hypothesis is to be definitively refuted.

Finally, if pituitary tumour-associated headache is indeed due to the production of a locally active nociceptive peptide, it is possible that CGRP and SP are the wrong candidate peptides. As mentioned in the general introduction, other neuropeptides that have been implicated in headache pathophysiology, and also play a role in paracrine activity of the hypothalamo-pituitary axis, including neuropeptide Y and vasoactive intestinal polypeptide (Jansen et al., 1994; Grouzmann et al., 1998; Hsu et al., 1989). Further studies are required to see if there is an association between these alternative peptides and headache in pituitary tumours.

In conclusion, although headache appears to be a significant cause of morbidity in patients presenting with pituitary tumours (Abe et al., 1998), particularly acromegaly and prolactinoma (Abe et al., 1998; Ezzat et al., 1994; Couch, 1986) there have been few studies regarding the mechanisms of headache. It has been discussed that severe headache may occur in patients with small functionally active tumours and Study 1 showed no relationship between tumour size and invasion with headache (Abe et al., 1998; Levy et al., 2004a). This has led to the hypothesis that headache may be caused

by the secretion of nociceptive peptides. This study suggests that CGRP and SP do not represent such peptides, although larger studies using molecular techniques may be required to confirm or refute these findings.

#### **4. Pituitary tumours and headache: clinical characteristics**

## **4.1 Study 3: phenotypic characteristics of pituitary tumour related headache**

Levy MJ, Matharu MS, Powell MP, Meeran K, Goadsby PJ

Abstract ref: Cephalalgia 2003; 23: 261

### **4.1.1 Abstract**

*Background:* Pituitary tumours may be associated with a variety of headache syndromes, although there are few prospective studies regarding the range of phenotypes observed.

*Methods:* Eighty-four patients with pituitary tumour related headache underwent a detailed interview regarding the clinical characteristics, associated features and response to treatment. The information was entered prospectively onto an electronic data base.

*Results:* The commonest phenotypic presentations were chronic (46%) and episodic (30%) migraine. Other headache syndromes included SUNCT (5%), cluster headache (4%), hemicrania continua (1%) and primary stabbing headache (27%). It was not possible to classify the headache according to International Headache Society diagnostic criteria in 6 cases (7%). Cavernous sinus invasion was present in the minority of presentations (21%), but was present in two of three patients with cluster headache. SUNCT-like headache was only seen in patients with acromegaly (n=2) and prolactinoma (n=2). Hypophysectomy improved headache in 49% and exacerbated headache in 15% of cases. Somatostatin analogues improved acromegaly-associated headache in 64% of cases, although rebound headache was described in three patients. Dopamine agonists improved headache in 25% and exacerbated headache in 21% of cases. In certain cases, severe exacerbations in headache were observed with dopamine agonists.

*Conclusion:* Headache appears to be a significant problem in pituitary disease and is associated with a range of phenotypes. The presenting phenotype is likely to be governed by a combination of factors including tumour activity, relationship to the cavernous sinus and patient predisposition to headache. A proposed modification of the current classification of pituitary-associated headache is given.



### **4.1.2 Introduction**

The clinical presentation of pituitary adenomas is dependent upon both structural and functional properties of the tumour (De Groot and Jameson, 2000). As discussed in Chapters 2 and 3, it is unclear whether headache, a common symptom of pituitary disease (Abe et al., 1998), is a structural or functional consequence of pituitary tumours. The presentation and mechanisms of headache in pituitary disease have not been widely investigated. Abe et al. (1998) described the headache characteristics in 19 patients with pituitary tumours, reporting generalised and predominantly bilateral frontal headache. However, with the advent of a systematic classification of headache (1988) and its subsequent revision (2004), the opportunity exists to carefully phenotype the headache seen with pituitary tumours. This effort allows the prospect of providing clinical information with which to manage such patients and may provide some insights into the primary headaches that are seen in pituitary disease. The aim of this study was to describe prospectively the phenotypic characteristics of pituitary tumour related headache in a large series of patients.

### **4.1.3 Subjects and Methods**

Eighty-four consecutive patients presenting with pituitary tumour related headache were studied between February 2001 and August 2003. An interview was conducted by a physician trained in headache (the candidate), during which a questionnaire was completed that required detailed documentation of headache characteristics and response to treatment. The information was entered prospectively onto an electronic data-base (Microsoft Access 2003).

#### *Headache*

Headache characteristics collected were laterality, site, severity, quality of pain, attack duration, frequency, and associated symptoms as well as triggers. I recorded response of the headache to surgery, radiotherapy and medical treatment. In each case, an attempt was made to classify the headache in line with the International Headache Society Diagnostic Criteria (1988), taking account ultimately of the revised second edition (2004).

### *Tumour*

Tumour size and the presence or absence of cavernous sinus invasion was also documented. Tumour size was classified according to maximum tumour diameter into the categories of micro-adenoma (< 1cm) and macro-adenoma (> 1cm). Cavernous sinus invasion was diagnosed on the basis on radiological appearance and treated as present or absent, and the laterality of cavernous sinus invasion was documented using standard radiological criteria (Cottier et al., 2000).

### *Disability*

Headache-related disability was assessed using a MIDAS questionnaire (Stewart et al., 2000).

## **4.1.4 Results**

### *Patient Demographics*

Of the 84 subjects interviewed, 60 were female (71%) and 24 male (29%). The mean age was  $44 \pm 1.4$  years (Table 17). The commonest tumour associated with headache was prolactinoma ( $n = 31$ ; 37%), followed by acromegaly ( $n = 28$ ; 33%), non-functioning adenoma ( $n = 20$ ; 24%), Cushings disease ( $n = 4$ ; 5%) and TSHoma ( $n = 1$ ; 1%). Full details are shown in Table 17.

### *Tumour characteristics*

For the whole group, 55 tumours were macro-adenomas (65%) and 29 were micro-adenomas (35%). Eighteen tumours (21%) were associated with cavernous sinus invasion. Macro-adenomas were commoner in the non-functioning adenoma (100%) and acromegaly groups (68%), whilst micro-adenomas were commoner in the Cushings (100%) and prolactinoma (52%) groups (Table 18).

**Table 17. Patient demographics and tumour characteristics**

Characteristic	Patients <i>n</i> (%)	
<i>Sex (n, %)</i>		
Female	60	(71)
Male	24	(29)
<i>Age (mean ± SD)</i>	44	± 1.4
<i>Tumour type (n, %)</i>		
Prolactinoma	31	(37)
Acromegaly	28	(33)
Non-functioning adenoma	20	(24)
Cushings disease	4	(5)
TSHoma	1	(1)
<i>Tumour characteristics</i>		
Macro-adenoma	55	(65)
Micro-adenoma	29	(35)
Cavernous sinus invasion	18	(21)

**Table 18. Tumour characteristics for each sub-type**

	<b>Macro-adenoma (n)</b>	<b>Micro-adenoma (n)</b>	<b>Cavernous sinus invasion (n)</b>
<b>Acromegaly (n = 28)</b>	19	9	7
<b>Prolactinoma (n = 31)</b>	15	16	6
<b>NFA (n = 20)</b>	20	0	4
<b>Cushings (n = 4)</b>	0	4	0
<b>TSHoma (n = 1)</b>	1	0	1
<b>Total (n = 84)</b>	55	29	18

## *Headache Characteristics*

### **Laterality**

Sixty patients (71%) had unilateral headache, which was side-locked in 53 (88%), while it was side variable in seven (12%; Table 19). Seventeen patients (20%) reported both bilateral and unilateral headache, whilst seven (12%) described exclusively bilateral symptoms. Of the 18 patients with cavernous sinus invasion, ten subjects (56%) experienced headache ipsilateral to the side of invasion.

### **Site**

The commonest location of headache was the orbital / retro-orbital (79%) and frontal (64%) region. Twenty-nine percent of patients had headache involving non-trigeminal territory and in 71% the trigeminal territory was exclusively involved. The full distribution of headache location is shown in Table 19.

### **Severity**

Ten patients (12%) graded their headache as moderate, 55 (65%) severe, 17 (20%) very severe, and 2 (2%) graded the pain excruciating. No patients graded their headache mild (Table 20).

### **Quality**

The range of pain quality is shown in Table 20. The commonest quality of pain was described as throbbing (63%).

### **Duration and frequency**

The median duration of headache exacerbation was seven hours (range 15 seconds – 96 hours; Table 21). The median attack frequency was 20 per month (range 1 – 30). Using the definition of Chronic Daily Headache as greater than 15 headache days per month (Welch and Goadsby, 2002) the frequency of chronic daily headache was 53%. Twenty-five patients (30%) used paracetamol (acetaminophen) - or codeine-containing agents on greater than 10 occasions per month, and were defined as having medication overuse.

## *Associated Symptoms*

The frequency and distribution of associated symptoms are shown in Table 21. The commonest associated symptoms were photophobia (71%) and nausea (58%). During an exacerbation, 64 patients (76%) preferred to lie still during an attack, 12 (14%) felt restless and preferred to move around, whilst 8 (10%) had no preference (Table 21).

Forty-two patients (50%) reported one or more cranial autonomic features in association with headache exacerbations (Table 21), the commonest of which were lacrimation (35%) and conjunctival injection (26%).

**Table 19. Headache laterality and site**

Characteristic	Patients <i>n</i> (%)	
<i>Laterality; n (%)</i>		
Strictly unilateral	60	(71)
Side-locked	53	(63)
Side-variable	7	(8)
Bilateral and unilateral	17	(20)
Strictly bilateral	7	(8)
<i>Site; n (%)</i>		
Orbital / retro-orbital	66	(79)
Frontal	54	(64)
Temple	30	(36)
Parietal	19	(23)
Vertex	26	(31)
Occiput	24	(29)
Nasal	4	(5)
Cheek	4	(5)
Teeth	4	(5)
Jaw	3	(4)
Ear	3	(4)
Neck	4	(5)

**Table 20. Headache severity and quality**

Characteristic	Patients <i>n</i> (%)	
<i>Severity; n (%)</i>		
Mild	0	(0)
Moderate	10	(12)
Severe	55	(65)
Very Severe	17	(20)
Excruciating	2	(2)
<i>Quality; n (%)</i>		
Throbbing	53	(63)
Sharp	29	(35)
Dull	21	(25)
Pressure	19	(23)
Stabbing	15	(18)
Tightening	13	(15)
Boring	6	(7)
Burning	3	(4)
Aching	3	(4)



**Table 21. Headache duration, frequency and associated features**

Characteristic	Patients <i>n</i> (%)	
<i>Median Duration (range)</i>	7	(15 sec – 96 hrs)
<i>Median attack frequency per month (range)</i>	20	(1 – 30)
<i>Chronic daily headache; n (%)</i>	45	(53)
<i>Analgesia overuse; n (%)</i>	25	(30)
<i>Associated symptoms</i>		
Nausea	49	(58)
Vomiting	18	(21)
Photophobia	60	(71)
Osmophobia	20	(24)
Restlessness	12	(14)
Aggravation with movement	64	(76)
<i>Cranial autonomic symptoms; n (%)</i>		
Ptosis	16	(19)
Eyelid oedema	10	(12)
Conjunctival injection	22	(26)
Lacrimation	29	(35)
Nasal blockage	10	(12)
Rhinorrhoea	6	(7)
Facial sweating	6	(7)
Facial flushing	2	(2)

## Triggers

The frequency and distribution of headache triggers are shown in Table 22.

## *Family history*

Forty-one patients (49%) reported a family history of a headache disorder (Table 22).

**Table 22. Headache triggers and family history**

Characteristic	Patients n (%)	
<i>Triggers; n (%)</i>		
Stress	54	(64)
Exertion	22	(26)
Hunger	34	(40)
Alcohol	22	(26)
Bright lights	15	(18)
<i>Family history of headache disorder; n (%)</i>	41	(49)

*International Headache Society (I.H.S) Classification (Headache Classification Committee of The International Headache Society, 2004)*

There were broadly two groups of patient diagnoses: those with phenotypes that mapped well onto accepted primary IHS diagnoses ( $n = 73$ ) and those that did not ( $n = 11$ ).

Of the former group, the commonest diagnosis was chronic migraine ( $n = 39$ ), followed by episodic migraine ( $n = 25$ ). Other headache diagnoses included Short-lasting Unilateral Neuralgiform headache attacks with Conjunctival injection and Tearing (SUNCT;  $n = 4$ ), cluster headache ( $n = 3$ ), hemicrania continua ( $n = 1$ ) and isolated primary stabbing headache ( $n = 1$ ). Twenty-two patients (26%) had primary stabbing headache as a second headache diagnosis (Table 23). The IHS headache diagnoses within each tumour sub-type are shown in Table 24. SUNCT syndrome was only seen in patients with prolactin- and growth hormone- secreting tumours, and primary stabbing headache was also more common in these two groups (87%).

Of those that did not map well onto an accepted primary IHS headache diagnosis, three patients were compatible with the current criteria for headache attributed to pituitary disease (IHS 7.4.4; Table 25). Of the remaining eight patients, two patients experienced featureless headache, which did not fit with tension-type headache or IHS 7.4.4 because the severity interfered with daily activities and the timing of headache was associated with the onset of pituitary disease but the treatment of the tumour did not absolutely resolve the headache (Criteria D; see Table 27). In 6 patients, it was not possible to classify the headache phenotype in accordance with the IHS criteria (Table 25). These patients had a mixture of migrainous (throbbing/nausea/photophobia/phonophobia) or cranial autonomic symptoms. These patients stood out in our groups experience with the particular mixture of symptoms and may represent a unique headache type seen in association with pituitary tumours.

**Table 23. Headache characteristics**

Characteristic	Patients <i>n</i> (%)	
<i>IHS Classification</i>		
Chronic migraine	39	(46)
Episodic migraine	25	(30)
SUNCT	4	(5)
Cluster headache	3	(4)
Hemicrania continua	1	(1)
Primary stabbing headache†	23	(27)
Other ††	11	(13)

† Lone diagnosis in one patient

†† Not definable by IHS criteria (2004), see Table 24

SUNCT Short-lasting Unilateral Neuralgiform headache, Conjunctival injection and Tearing

**Table 24. International Headache Society (I.H.S) Classification (2004) diagnoses for each tumour subtype**

	<b>CM</b>	<b>EM</b>	<b>SUNCT</b>	<b>CH</b>	<b>HC</b>	<b>PSH</b>	<b>Other†</b>
<b>Acromegaly (n = 28)</b>	15	6	2	0	0	8	3
<b>Prolactinoma (n = 31)</b>	11	13	2	1	1	12	4
<b>NFA (n = 20)</b>	12	4	0	2	0	3	3
<b>Cushings (n = 4)</b>	1	3	0	0	0	0	1
<b>TSHoma (n = 1)</b>	1	0	0	0	0	0	0
<b>Total (n = 84)</b>	40	26	4	3	1	23	11

CM            Chronic migraine

EM            Episodic migraine

SUNCT       See Table 7

CH            Cluster headache

HC            Hemicrania continua

PSH          Primary Stabbing Headache

NFA          Non-functioning adenoma

†            Not definable by IHS criteria

**Table 25 Patients with potentially secondary headache phenotypes (Headache Classification Committee of The International Headache Society, 2004)**

Case	Age/ Sex	Effect of surgery	Tumour Characteristics			Headache Characteristics							
			Histology	Size	Cavernous sinus invasion	Site	Quality	Severity	Mvt	Nausea	Photo	Phono	Cranial autonomic
<b>Featureless Headache phenotypes</b>													
GS	74/M	Better	NFA	Macro	-	Orbital/retro-orbital	Continuous Dull	Moderate	-	-	-	-	-
GW	84/F	Better	NFA	Macro	-	Retro-orbital/vertex	Continuous Pressure	Moderate	-	-	-	-	-
PC	65/M	Medical Rx only better	Prolactin	Macro	Right	Strict right	Orbital/parietal/ vertex Tight/sharp 2 hr stabbing exacerbations	Severe	-	-	-	-	-
NE	31/F	Worse	Prolactin	Macro	-	Frontal	Episodic 3 hours Dull	Moderate	-	-	-	-	-
MN	37/F	Worse	Acro	Macro	Left	Strict left retro-orbital	Continuous dull ache	Severe	-	-	-	-	-
<b>Headaches with mixed migraine/trigeminal autonomic cephalalgia phenotypes</b>													
LD	29/F	Better	Acro	Macro	Left	Strict left retro-orbital/frontal	Continuous pressure plus stabs	Severe	+	-	+	+	Lacrimation
IT	50/M	Better	Acro	Macro	-	Uni/Bilateral Temporal/vertex	Ache/boring/dull	Very severe	+	+	+	-	Lacrimation
SB	38/M	Better	Prolactin	Macro	Left	Strict left	Continuous	Severe	R	-	+	-	Lacrimation

						Orbital/retro-orbital	boring/dull 30-60 exacerbations						ptosis
PT	52/F	Better	NFA	Macro	-	Bilateral frontal	Continuous dull/throbbing 60 min exacerbations ± visual aura	Severe	R	-	-	-	Lacrimation
CW	51/F	Better	Cushings	Micro	-	Bilateral generalised	Episodic (4-48 hrs) pressure	Severe	-	-	-	+	-
BP	73/M	Worse	Prolactin	Macro	Left	Strict Left Orbital/maxilla /jaw	Continuous Sharp Plus stabs	Very severe	-	+	+	-	Nasal block ptosis



## *Treatment characteristics*

### **Surgery**

Fifty-five patients (65%) underwent hypophysectomy, of which 50 had transphenoidal, and 5 trans-cranial approaches. Twenty-seven patients (49%) reported an improvement in headache following surgery, 20 (36%) experienced no change in symptoms, and 8 (15%) reported worsening of headache (Table 26).

### **Radiotherapy**

Sixteen patients underwent radiotherapy, of which one experienced an improvement in headache, the remainder reporting no change in symptoms (Table 26). The median time to follow up from radiotherapy was 5 years (range 2 – 13 years).

### **Somatostatin analogues**

#### **Octreotide**

Twelve patients received octreotide 100µg, of which seven reported a reduction in headache frequency and severity (Table 26). Of those that experienced improvement in headache, four experienced improved migraine, two reported an improvement in featureless headache, and one reported a reduction in the frequency and severity of SUNCT-like attacks. After several months of octreotide administration, three patients experienced rebound headache. One patient developed a dependency syndrome, requiring twelve injections per day. Four patients received both octreotide and lanreotide during treatment, three of who reported a preferential response to octreotide in terms of headache.

#### **Octreotide LAR**

Six patients received octreotide LAR 20mg per month, of which four reported a reduction in headache frequency and severity (Table 26). Two patients reported headache recurrence one week prior to the following injection. No patient on octreotide LAR developed tachyphylaxis or a dependency syndrome.

#### **Lanreotide**

Four patients received lanreotide 30mg every two weeks. One patient experienced reduction in headache frequency and severity on lanreotide, the remaining three reporting no change in symptoms (Table 26). The single patient who experienced improvement in headache on lanreotide did not experience benefit from octreotide.

#### Dopamine agonists

##### Cabergoline

Cabergoline (dose range 0.25 – 4mg per week) was prescribed in 23 patients. Nine patients reported a reduction in headache severity and frequency on cabergoline, 11 experienced no change, and 3 reported an exacerbation in symptoms (Table 26). Of the 3 patients who reported an exacerbation in symptoms, one experienced a change from episodic to chronic migraine, one underwent a change from episodic migraine to persistent unilateral indomethacin-responsive headache, classified as hemicrania continua, and one experienced a severe and reproducible exacerbation of SUNCT-like syndrome that lasted for 12 hours.

##### Bromocriptine

Twenty-two patients received bromocriptine (dose range 2.5 – 22.5 mg per day). Of these, three patients experienced a reduction in headache frequency and severity, 13 reported no change, and six reported headache exacerbation (Table 26). Of the six patients that reported headache exacerbation, five experienced worsening migraine, and one experienced severe exacerbation of SUNCT lasting 12 hours.

##### Quinagolide

Two patients received quinagolide therapy. One experienced worsening migraine whilst the other reported an exacerbation in SUNCT identical to the cabergoline and bromocriptine responses described above (Table 26).

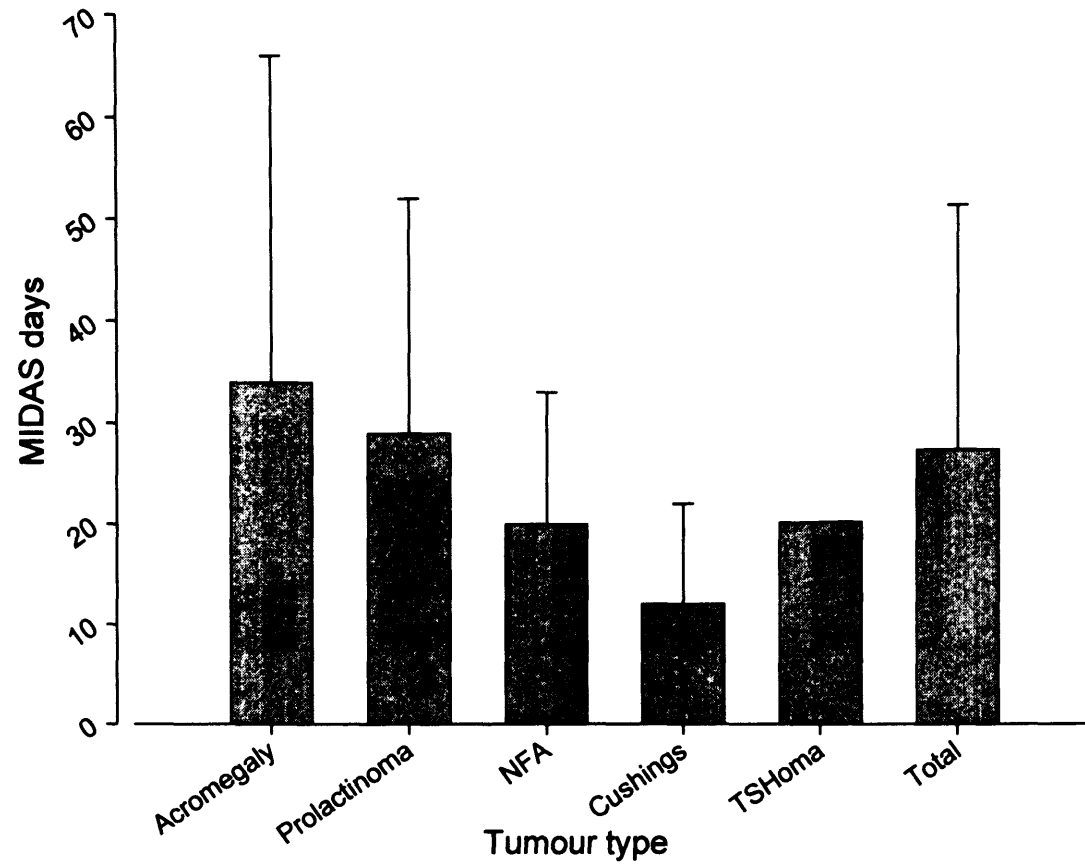
**Table 26 Response of headache to treatment of pituitary disease**

	<b>Improvement</b>	<b>Exacerbation</b>	<b>No change</b>
<b>Surgery</b>			
Transphenoidal	23	8	19
Transcranial	4	0	1
<b>Radiotherapy</b>	1	0	15
<b>Somatostatin analogue</b>			
Sandostatin	7	0	5
Octreotide LAR	4	0	2
Lanreotide	1	0	3
<b>Dopamine agonist</b>			
Cabergoline	9	3	11
Bromocriptine	3	6	13
Quinagolide	0	2	0

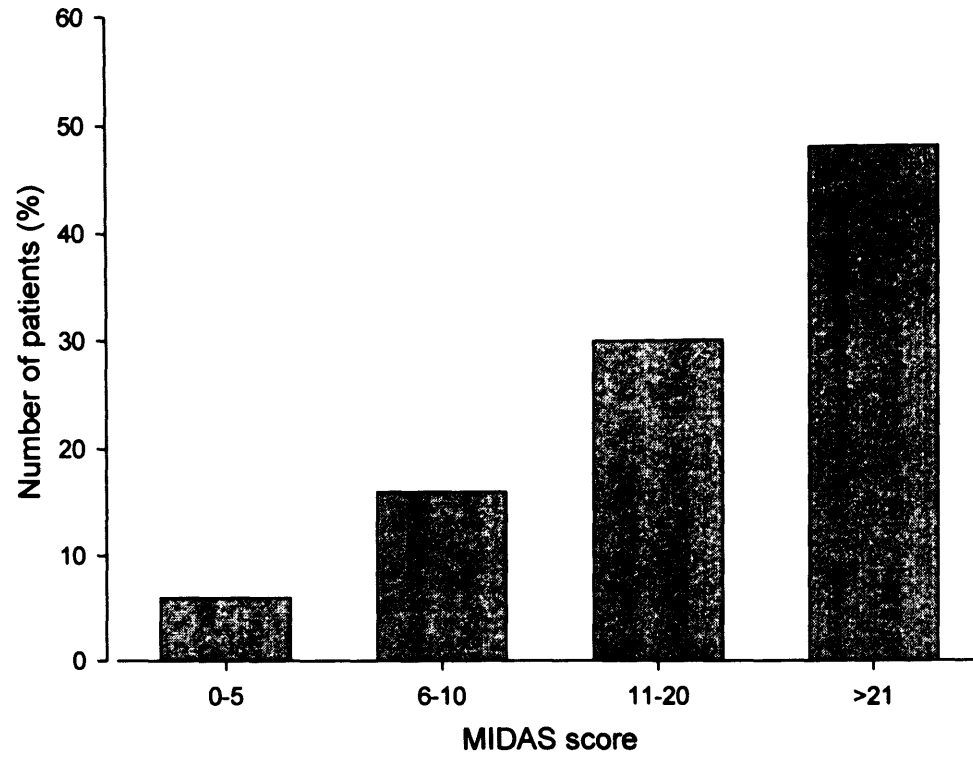
## Quality of life

MIDAS questionnaires were completed in 69 patients. The highest MIDAS scores were seen in the acromegaly and prolactinoma groups (Figure 15). The mean MIDAS score for the whole group was  $27 \pm 24$  days. Forty-eight percent of patients with pituitary tumour associated headache had severe levels of disability (Figure 16)

**Figure 15 Distribution of MIDAS scores amongst tumour types with the mean  $\pm$  SD shown for each tumour and for the group as a whole (total)**



**Figure 16 Distribution of MIDAS scores by conventional grading cut-offs in patients with pituitary tumour and headache. The distribution is right-shifted in comparison to primary headache.**



#### **4.1.5 Discussion**

Headache is a common and disabling aspect of pituitary disease. The cohort most often reported migraine, but cluster headache, Short-lasting Unilateral Neuralgiform headache attacks with Conjunctival injection and Tearing (SUNCT) and hemicrania continua was also observed. Some patients had unclassifiable headaches, which may be new forms of secondary headache specific to pituitary tumours that have not hitherto been recognised. No particular tumour type produced a specific headache syndrome. The observation that 48% of patients had MIDAS scores within the severe range suggests that disability due to pituitary headache is considerable (Lipton et al., 2001). Pituitary tumour-related headache is an important, common issue that requires careful history to facilitate correct diagnosis and thus specific management of those afflicted.

The improvement in headache following surgery in 49% of cases implies a causal link between the tumour and presence of headache, although it is difficult to control for the confounding variables of the anaesthetic, or indeed natural history. Furthermore, the abolition of headache in 64% of acromegalics who were prescribed somatostatin analogues suggests a link between tumour activity and headache. Octreotide appeared to be more beneficial than lanreotide for headache, although one patient responded preferentially to lanreotide. As discussed in Section 1.15.2, it is possible that the somatostatin receptor status of the tumour is important in predicting headache response. Some patients in the cohort who had migraine headache reported a useful therapeutic effect with octreotide, and an interesting question is whether this drug is useful in non-pituitary tumour headache, which is investigated in Chapter 6. Octreotide dependency has been previously reported (Popovic et al., 1988, May et al., 1994) and this was observed in the cohort as a potential complication in the management of pituitary-related headache.

Dopamine agonists both alleviated and exacerbated headache, which has previously been observed (Ferrari et al., 1988, Massiou et al., 2002) and described in the Appendix (Cases 1, 2 and 4). This paradoxical observation may be related to a complex interplay of the physical effects on the tumour and the central actions of dopamine agonists. The reduction of tumour size in large prolactinomas may improve

headache via structural changes, although the data from Study 1 and others (Abe et al., 1998) suggests that tumour size does not correlate with headache. Alternatively, or in addition, the effects of dopamine agonists on the trigeminovascular system may have deleterious effects on headache. Dopamine agonists share properties with ergot alkaloids (Trabucchi et al., 1978), and ergot alkaloids are known to alter the activity of the trigeminovascular system (Hoskin et al., 1996). As discussed in Sections 1.4 and 1.5, it has also been suggested that the dopamine-prolactin axis plays an important role in some forms of primary headache, notably migraine (Peroutka et al., 1997; Peres et al., 2001) and cluster headache (Goadsby, 2002). This may, in part, explain the unpredictable headache responses observed with dopamine agonists. The exacerbation of headache was dramatic in certain cases, an observation that has been described in Appendix Cases 1 and 2, and previously observed in association with SUNCT (Ferrari et al., 1988, Massiou et al., 2002).

In addition to tumour-related factors, the type of headache in pituitary disease is likely to be a result of patient-dependent factors. The finding that 49% of the study group had a family history of headache suggests that they were more predisposed to the primary headaches than the general population (Steiner et al, 2003). As discussed in Section 1.4, migraine is known to have a familial aggregation (Ferrari, 1998) and the development of pituitary tumour-associated migraine, accounting for 75% of presentations in this study, may have been a result of genetic predisposition to migraine in affected patients rather than specific tumour-related factors. As migraineurs have an increased sensitivity to any external changes in the internal or external *milieu* compared to controls (Goadsby et al., 2002) it may be that the development of the pituitary tumour simply lowered the threshold for attacks in predisposed migraineurs. The presence of a higher proportion of migraine in prolactinomas and growth hormone-secreting tumours suggests that functional activity may be an important trigger.

Cluster headache and SUNCT are relatively rare headache syndromes and the observation of three cases of cluster headache and four cases of SUNCT in this relatively small cohort of 84 patients suggests that these phenotypes may be over-represented in pituitary disease. While it is possible that this is in part referral bias with regard to the unit's headache interest where I conducted the thesis (QS), this was



minimised by studying consecutive referrals to the neurosurgery unit, which is unlikely to have this headache-related bias. Cavernous sinus invasion was present in two of the three cluster cases which may suggest that invasion of local structures is relevant to this headache syndrome. Although Study 1 showed that the cavernous sinus invasion is not predictive of headache in pituitary tumours, *per se* (Abe et al., 1998; Levy et al., 2004a), the sinus does contain pain-producing structures, such as the internal carotid artery and trigeminal nerve and ganglion, invasion of which might be expected to cause pain. As discussed in Section 1.8.3, there have been several reports of pituitary-associated cluster headache presenting with ipsilateral cavernous sinus tumour invasion (Tfelt-Hansen et al., 1982; Greve a& Mai, 1988; Milos et al., 1996; Porta-Etessam et al., 2001). The cavernous sinus has been previously implicated in the pathophysiology of cluster headache (Moskowitz, 1988; Harbedo, 1994), although functional imaging data suggests that ipsilateral hypothalamic activation may be more important (May et al., 1998a, May et al., 2000; Sprenger et al., 2004). Of the four SUNCT cases, two were prolactinomas and two were growth hormone-secreting tumours, suggesting that tumour activity may be important in pituitary-related SUNCT, although our sample size is small. The dramatic exacerbation of SUNCT with dopamine agonists observed in certain cases further suggests that perturbations in the dopamine-prolactin axis may be important in this headache syndrome. As described in Section 1.5.2, ipsilateral hypothalamic activation has been demonstrated in primary SUNCT (May et al, 1999) and it is conceivable that specific neuroendocrine pathways involving the dopamine-prolactin and growth hormone axis are capable of activating SUNCT pathophysiology.

The aim of this study was to document the clinical spectrum of pituitary tumour-associated headache. No attempt was made to determine the prevalence of headache in pituitary disease, which would have required recruitment of larger numbers of patients from both the surgical and non-surgical setting in a prospective and consecutive fashion. Because the study was based in a neurosurgical centre, the patient population is likely to have contained relatively larger numbers of macroadenomas compared to a non-surgical centre. This may have given a biased impression of the frequency and quality of headache found in our study, and further work is required to determine the validity of the findings in the generality of patients with pituitary tumours. I observed a significant number of patients who experienced

residual headache after treatment of their pituitary tumour and found these patients to present a difficult management problem. Although treatment response was not formally part of the study design, a large number of this cohort was managed in a dedicated clinic. I observed that phenotype-driven medical management markedly improved the patients' disability. There are previous reports that pituitary tumour-associated with headache may respond to serotonin-5-HT<sub>1B/1D</sub> receptor agonists, triptans (Shah & Frei, 1999; Pascual 2000). I found that both acute and preventive phenotype-driven treatment was helpful for many patients. Patients with residual ipsilateral cavernous sinus invasion were particularly refractory to medical therapy. Prospective blinded placebo-controlled studies are required to determine the optimum management of the pituitary tumour-associated headache, although in their absence placebo-controlled studies from the underlying primary headache types manifest by these patients seem a very useful guide to their management.

Lastly, based on the relatively large, prospective study with tissue verification of the diagnosis, it may be possible to make some new suggestions as to the classification of headache in patients with pituitary disease (Table 27). Currently the International Headache Society classifies pituitary and hypothalamic headaches together (Headache Classification Committee of The International Headache Society, 2004). However, because pituitary adenomas are clinically distinct from hypothalamic syndromes, it seems reasonable to split these into separate entities. Despite the negative correlation between cavernous sinus invasion and headache found in Study 1, the implications of local involvement of the cavernous sinus from both a local treatment and headache presentation, suggest that it is sensible to specify the presence or absence of cavernous sinus invasion for research and characterisation (Table 27). It is suggested that requirement C (Table 27) is insufficient to deal with non-functioning adenomas, which made up nearly one quarter of the cohort, as it fails to demonstrate that the majority of these tumours present with a non-endocrine manifestation of the tumour, such as visual impairment. For requirement D (Table 27) the requirement of complete resolution of headache after surgical or endocrine management is not uniformly useful, as many patients were observed to have improvement in headache rather than complete resolution post-treatment. Therefore it seems appropriate that amelioration of headache after tumour treatment, rather than resolution more completely captures the outcome, thus it is suggested that section D is altered. With publication of this

study, it is hoped that other centres will consider these proposals and test them prior to the next edition of the headache classification.

**Table 27 Proposals for modifications to the I.H.S criteria**

***Current classification***

**7.4.4 Headache attributed to hypothalamic or pituitary hyper- or hyposecretion**

***Diagnostic criteria:***

- A. Bilateral, frontotemporal and/or retro-orbital headache fulfilling criteria C and D
- B. At least one of the following:
  - 1. prolactin, growth hormone (GH) and adrenocorticotrophic hormone (ACTH) hypersecretion associated with microadenomas <10 mm in diameter
  - 2. disorder of temperature regulation, abnormal emotional state, altered thirst and appetite and change in level of consciousness associated with hypothalamic tumour
- C. Headache develops during endocrine abnormality
- D. Headache resolves within 3 months after surgical resection or specific and effective medical therapy

***New proposal***

**7.4.4 Headache attributed to hypothalamic dysfunction**

***Diagnostic criteria:***

- A. Bilateral, frontotemporal and/or retro-orbital headache fulfilling criteria C and D.
- B. Disorder of temperature regulation, abnormal emotional state, altered thirst and appetite and change in level of consciousness associated with hypothalamic tumour
- C. Headache develops when hypothalamic pathology is manifest
- D. Headache resolves within 3 months after specific and effective medical therapy

**7.4.5 Headache attributed to pituitary disease**

***Diagnostic criteria:***

- A. Bilateral or unilateral frontotemporal and/or retro-orbital headache fulfilling criteria C and D
- B. Either a functioning or non-functioning pituitary tumour is identified by biochemical testing or appropriate brain imaging
  - a. with cavernous sinus involvement
  - b. without cavernous sinus involvement
- C. Headache develops in close temporal proximity to endocrine abnormality or with structural symptoms attributable to pituitary disease, such as visual loss
- D. Headache resolves, or there is marked improvement, within 3 months after surgical resection, or specific and effective medical therapy

In summary, I have described the headache characteristics observed in 84 patients with pituitary tumour-associated headache. Functioning tumours presented with the most headache-related disability, and the dopamine-prolactin and growth hormone axes were exclusively associated with SUNCT. The majority of cases of pituitary-associated headache presented with migraine, although a wide spectrum of headache presentations was observed. The current classification system is useful but suggestions for revision have been suggested based on the data. From a clinical perspective, pituitary-associated headache appears to be a management problem both pre-and post-treatment of the pituitary tumour and warrants further study to optimise outcome.

## **Somatostatin withdrawal and headache**

## **5.1. Study 4: Somatostatin infusion withdrawal: a study of patients with migraine, cluster headache and healthy volunteers**

Levy MJ, Matharu MS, Lightman SL, Goadsby PJ

Paper ref: Pain 2003; 102 (32): 237-242

### **5.1.2 Abstract**

*Background:* Migraine and cluster headache are the most common disabling primary headache syndromes and are typically episodic. A reliable method of triggering such headache attacks facilitates the study and treatment of these disorders. There is sufficient clinical and laboratory evidence to suggest that somatostatin withdrawal may be a useful way of triggering headache.

*Methods:* 15 subjects were studied; 8 migraineurs, 4 cluster headache sufferers and 3 healthy controls. Each subject had a standard somatostatin infusion, 250µg/h for 3.5 hrs. Subjects were followed for 24 hours post-infusion to observe headache response following cessation of infusion.

*Results:* Growth hormone was suppressed in each subject demonstrating a biologically active infusion of somatostatin. None of the non-headache sufferers had pain. Seven of eight migraine sufferers had no immediate headache and no delayed headache. One migraineur experienced short lasting headache with no migrainous features. Three of four patients with cluster headache had no significant pain with the infusion, while one had pain after one hour.

*Conclusion:* The results suggest that somatostatin infusion is not a reliable way to produce headache in experimental settings in either migraine or cluster headache. The data do not exclude a role for somatostatinergic mechanisms in primary headache.

### 5.1.3 Introduction

This chapter utilises the observations that somatostatinergic mechanisms may be important in headache. It has been observed in the previous chapter that overuse of octreotide in patients with pituitary tumours may give rise to increased headache. An interesting question is whether over-exposure to somatostatin may trigger headache in patients with primary headache. Migraine and cluster headache are the most common disabling primary headache syndromes. In their typical form a key feature of both disorders is episodicity (Headache Classification Committee of The International Headache Society, 1988). This feature makes the study of these conditions particularly challenging. A reliable method of triggering either condition would be, at least, extremely useful and may provide insight into the disorder in terms of development of new acute attack or preventative treatment strategies.

Nitroglycerin (Thomsen et al., 1994) and histamine (Krabbe et al., 1980; Lassen et al., 1995) have both been used to induce migraine and cluster headache attacks (Ekbom et al., 1968). Patients with migraine develop symptoms usually several hours after exposure to nitroglycerin (Iversen, 2001) whilst cluster patients (during a cluster period) may have an attack within minutes (Ekbom et al., 1968). A problem with these existing models is that the most reliable and best studied model, that of nitroglycerin-triggering, is directly associated with vasodilatation (Thomsen et al., 1994) and thus dissecting the non-vascular mechanisms in these headache types is a considerable challenge. It would thus be desirable to have a non-vasoactive method for triggering migraine or cluster headache, or both.

The evidence that somatostatin plays an important role in pain modulation has been reviewed in the introduction to the thesis. It has been observed that somatostatin, after repeated or prolonged exposure, may be pro-nociceptive (Popovic et al., 1988) while repeated exposure to somatostatin analogues may lead to a dependency syndrome, which is characterised by headache induction and rebound headache (May et al., 1994). Similarly, cluster headache has been reported to be induced by a somatostatin analogue (Otsuka et al., 1998). The administration of native somatostatin to healthy individuals causes suppression of growth hormone, which rebounds when it is discontinued. Endocrinologists have observed that patients may experience headache



after somatostatin withdrawal during metabolic studies (S Lightman, unpublished observations). I aimed to study systematically the effect of somatostatin withdrawal after prolonged intravenous exposure in patients with migraine, cluster headache and healthy controls. Headache scores were recorded before, during and after somatostatin infusion, in order to determine any correlation between headache and serum growth hormone, in an attempt to characterise a new model for triggering primary neurovascular headache.

#### **5.1.4 Subjects and Methods**

##### *Patients*

Fifteen subjects were studied (8 migraine, 4 cluster headache and 3 healthy controls). Headache diagnoses were based on International Headache Society Diagnostic Criteria (Headache Classification Committee of The International Headache Society, 2004). Patients with cluster headache were recruited from the headache clinic (QS). Migraineurs and healthy controls were recruited both from the headache clinic and via local advertisements. The episodic cluster headache patients included in the study were in the middle of their bout. All had attacks in the weeks following the trial confirming that they were still in the bout at the time of the infusion. None of the patients were on preventative medication. Exclusion criteria included diabetes mellitus and ischaemic heart disease. The study was approved by the local ethics committee (Study Ref: 01 / N062).

##### *Study Design*

Patients arrived fasted and an intravenous cannula was inserted in the antecubital vein of each forearm. Native somatostatin (Stilamin ®, Serono) was infused at a rate of 250µg/h for 3 ½ hours, according to previously published protocols (Villaume et al., 1986). Subjects remained awake and supine throughout the procedure. Serum glucose was monitored at 10-minute intervals for the first 30 minutes of the study. No hypoglycaemic episodes were recorded and no adverse effects were observed during or after somatostatin infusion. A headache visual analogue (VAS) score was taken at 15-minute intervals throughout the study. Serum growth hormone (GH) was assayed at 0, 60, 120 and 180 minutes. After somatostatin infusion withdrawal, the VAS score was taken at 5 minute intervals and serum GH taken at 5, 15 and 30 minutes post-

infusion. Subjects subsequently kept a headache diary for 24 hours and were followed up to see if headache occurred after discharge from hospital.

### *Statistical considerations*

The study was designed as a pilot one to consider the utility of somatostatin infusion withdrawal in triggering migraine or cluster headache. As such the study was powered based on the most reliable method of triggering migraine and cluster headache, which is by administration of a nitric oxide donor. The nitroglycerin migraine model produces migraine attacks in 90% of patients (Iversen, 2001) while histamine infusion, which also acts as an NO donor, produces headache in 100% of patients (Lassen et al., 1995). It was considered that any new model would need to trigger attacks in at least 80% of patients to be worth further characterisation. It was thus calculated that for 8 patients the study would have an 80% power to detect a difference from the NO trigger if just one were to have migraine with a  $P < 0.05$ , using a chi-square test. For cluster headache, nitroglycerine has long been known to be a very reliable trigger (Ekbom, 1968). It has been shown to trigger 85% of patients in their bout to have headache (Matharu and Goadsby, unpublished data). It was considered that any new model system that consecutively failed in four patients was unlikely to be helpful.

### **5.1.5 Results**

Each subject completed the acute phase of the study and was followed up for at least 24 hours to determine if any delayed headache had been triggered. The patient demographics and headache responses are outlined in Table 28. Demonstrating that a biologically active infusion of somatostatin had been employed, growth hormone was appropriately suppressed in all subject groups (Table 29, Figures 17 – 20). There was no difference in growth hormone response in all three groups (Figure 20).

#### *Control Group*

The control group, volunteers without headache ( $n = 3$ ) did not develop any significant headache symptoms before or after the somatostatin infusion (Figure 21).

#### *Migraine Group*

In the migraine group, 7 of 8 patients had neither an acute headache nor did they experience any form of delayed headache. One patient in the migraine group developed a mild/ moderate unilateral headache during the infusion that remained mild for three hours after the infusion was withdrawn and subsequently subsided (Figure 22). This headache had no migrainous features.

#### *Cluster Headache Group*

Of the cluster headache patients, 3 of 4 patients had no immediate nor any delayed attacks after the somatostatin infusion. One patient in the cluster group developed an attack during the first hour of the infusion (Figure 23). This was aborted with 100% oxygen within ten minutes and did not return. One patient had an attack at 2am, 17 hours after the infusion that was in keeping with his typical attack periodicity.

There was no correlation between headache score and growth hormone for all three groups (Pearson correlation 0.91;  $p=0.357$ : Figure 24)

**Table 28. Patient demographics and headache response to somatostatin infusion**

Diagnosis*	Sex	Age	Headache Score (0-10) with somatostatin infusion						
			0min	60min	120min	180min	+5min	+15min	+30min
Migraine with aura	M	40	0	2	1	0	0	0	0
	F	29	0	0	0	0	0	0	0
Migraine without aura	M	24	0	1	0	0	1	0	0
	F	41	0	0	0	1	0	0	0
	F	37	0	0	0	0	0	0	0
	F	37	0	0	1	0	0	0	0
	M	44	2	4	5	5	5	5	5
	F	39	0	1	0	0	1	0	0
Chronic cluster	M	53	0	7	0	0	0	0	0
	M	47	2	3	3	4	4	0	0
Episodic cluster	M	36	0	0	0	0	0	0	0
	M	33	0	0	0	0	0	0	0
Controls	M	32	0	0	0	0	0	0	0
	M	33	0	0	0	0	0	0	1
	M	54	0	0	0	0	0	0	0

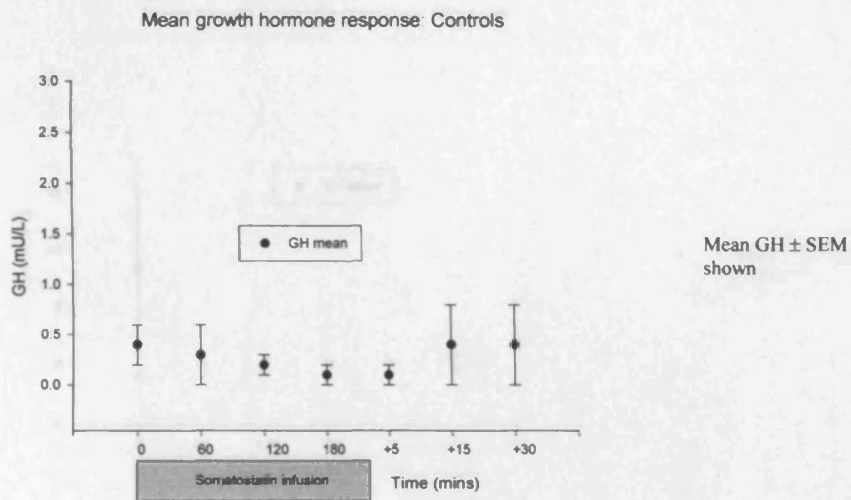
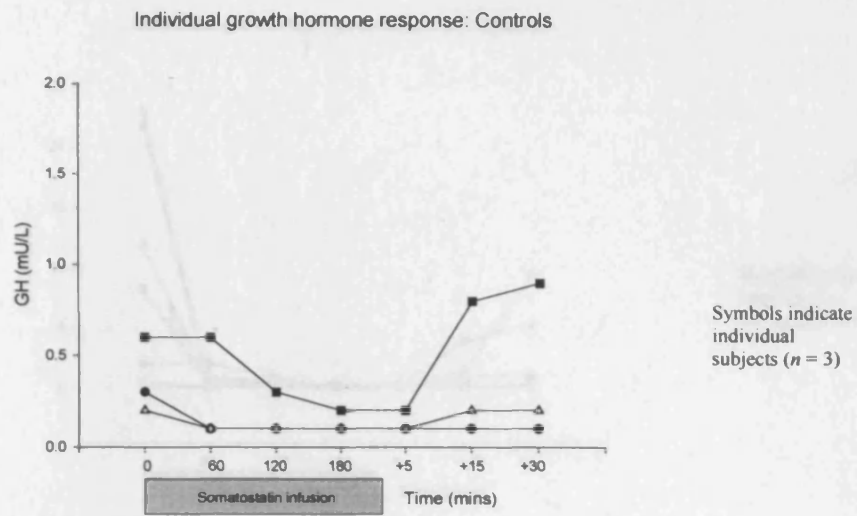
\*Clinical phenotype according to International Headache Society (1988)

**Table 29. GH response to somatostatin infusion**

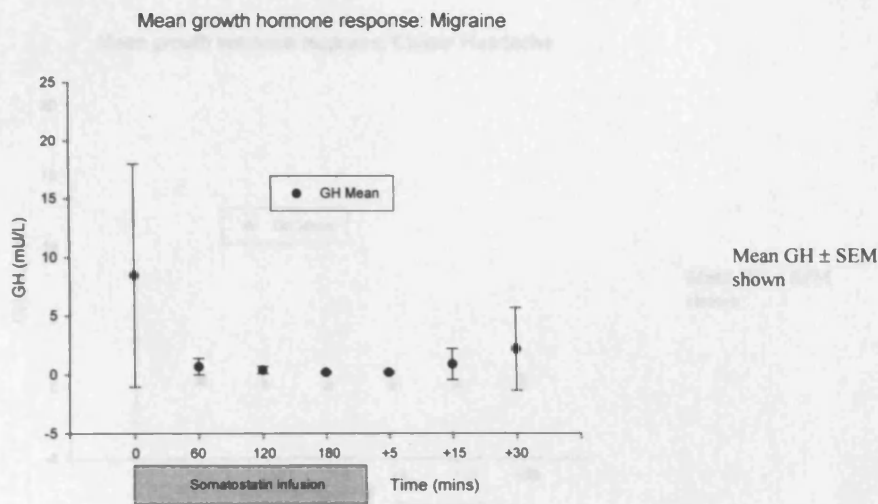
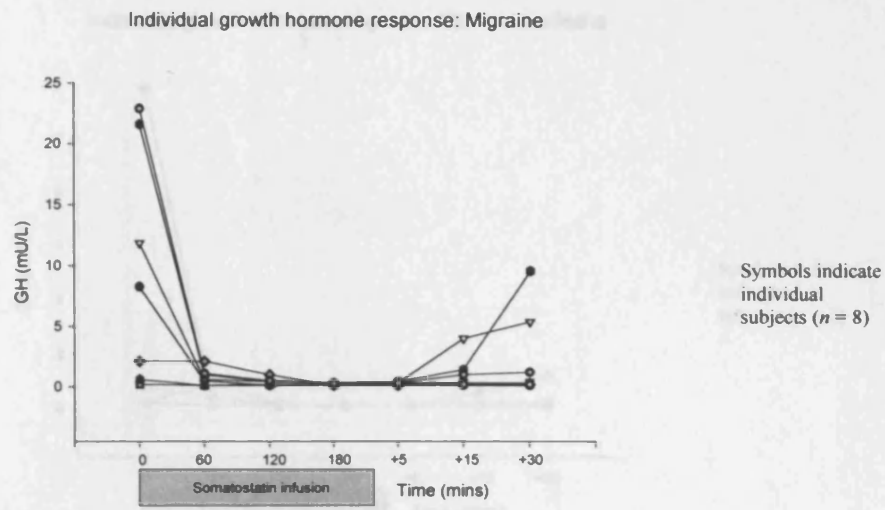
<b>Diagnosis*</b>	<b>Growth Hormone response to somatostatin (mU/L)</b>						
	<b>0 min</b>	<b>60min</b>	<b>120 min</b>	<b>180min</b>	<b>+5min</b>	<b>+15min</b>	<b>+30min</b>
<b>Migraine with aura</b>	8.3	0.5	0.1	0.1	0.1	0.1	0.1
	0.2	0.1	0.2	0.2	0.2	0.3	0.3
<b>Migraine without aura</b>	0.6	0.1	0.1	0.1	0.3	0.3	0.3
	11.9	0.6	0.4	0.3	0.3	4.0	5.4
	0.6	0.1	0.1	0.1	0.1	0.2	0.2
	21.6	1.0	0.4	0.3	0.4	1.4	9.6
	2.1	2.1	1.0	0.1	0.1	0.2	0.2
	22.9	1.1	0.5	0.3	0.3	1.0	1.2
<b>Chronic cluster</b>	0.8	0.4	0.2	0.1	0.1	0.1	0.1
<b>Episodic cluster</b>	0.2	0.9	0.2	0.1	0.1	0.2	0.2
	0.2	0.1	0.1	0.1	0.1	0.6	1.2
	11.9	0.4	0.1	0.1	0.1	0.2	0.2
<b>Controls</b>	3.1	0.1	0.1	0.1	0.1	0.1	0.1
	0.6	0.6	0.3	0.2	0.2	0.8	0.9
	0.2	0.1	0.1	0.1	0.1	0.2	0.2

\*Clinical phenotype according to International Headache Society (1988)

**Figure 17. Effect of somatostatin on GH. Control Group**

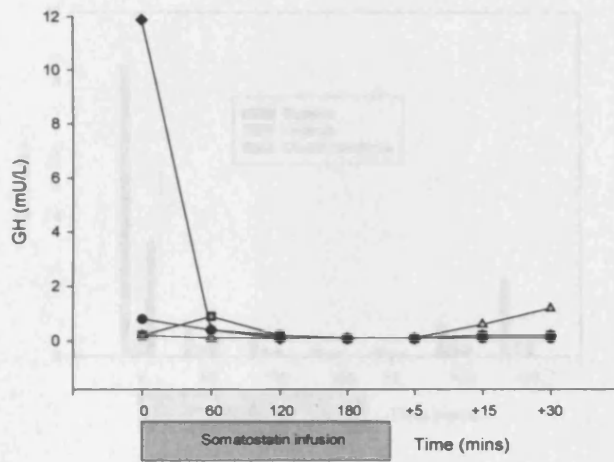


**Figure 18. Effect of somatostatin on GH. Migraine Group.**

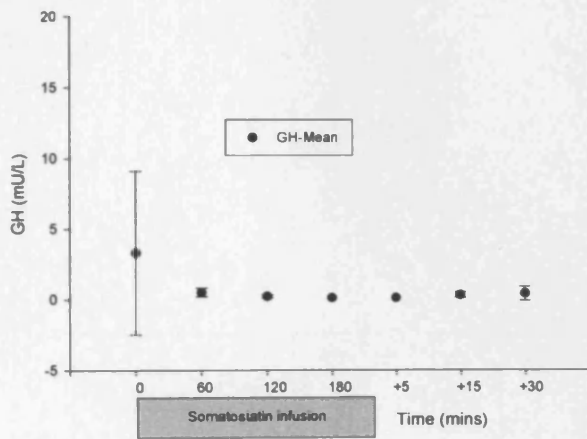


**Figure 19. Effect of somatostatin on GH. Cluster Group.**

Individual growth hormone response: Cluster Headache

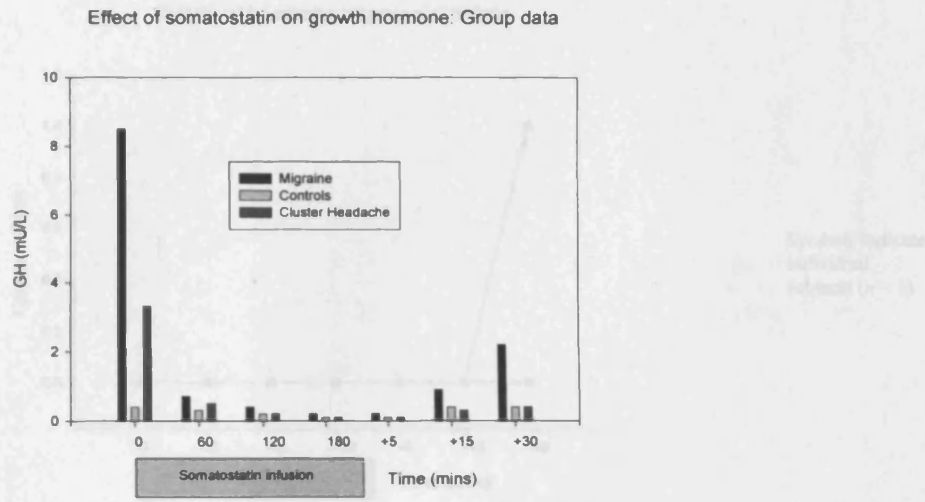


Mean growth hormone response: Cluster Headache

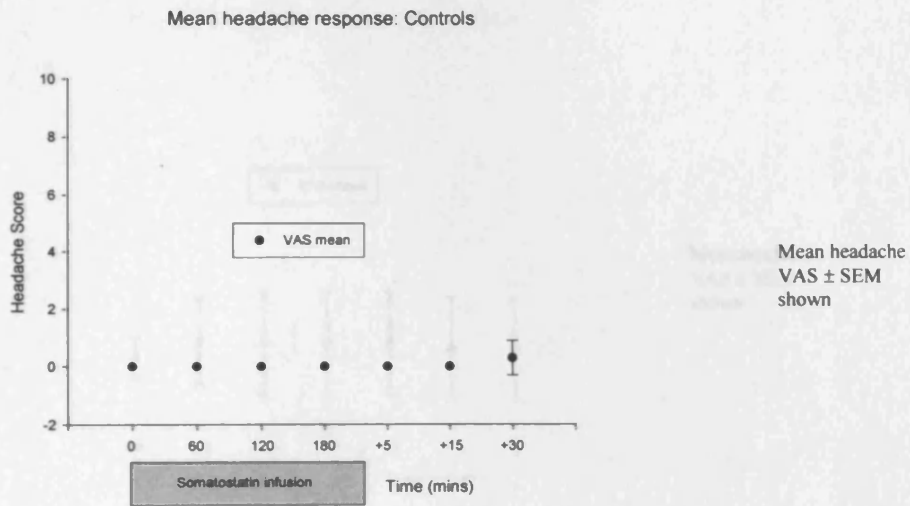
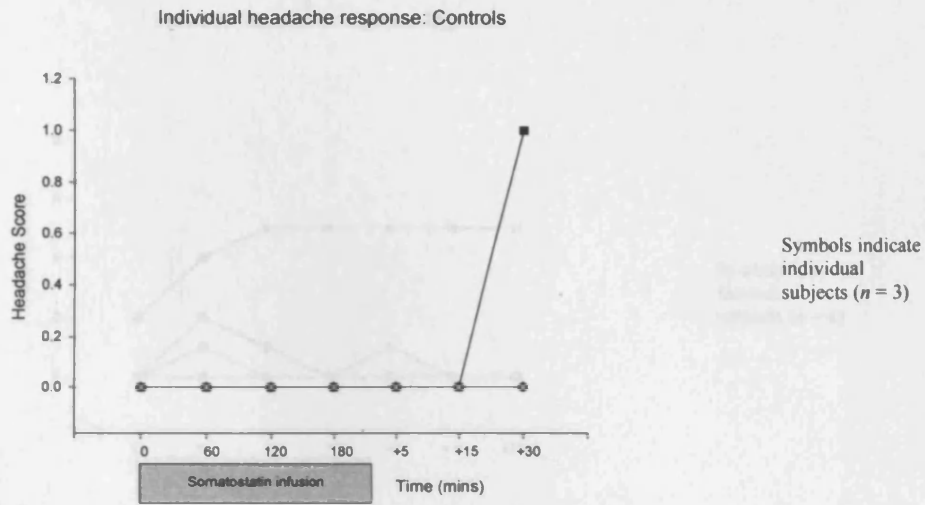




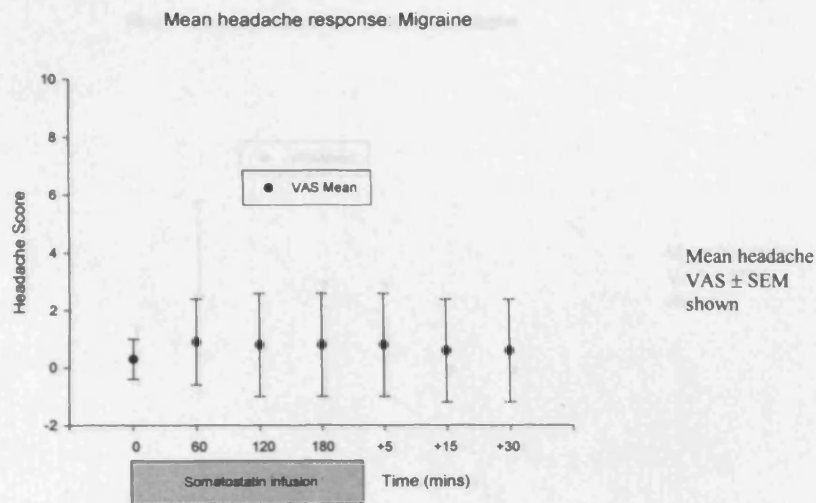
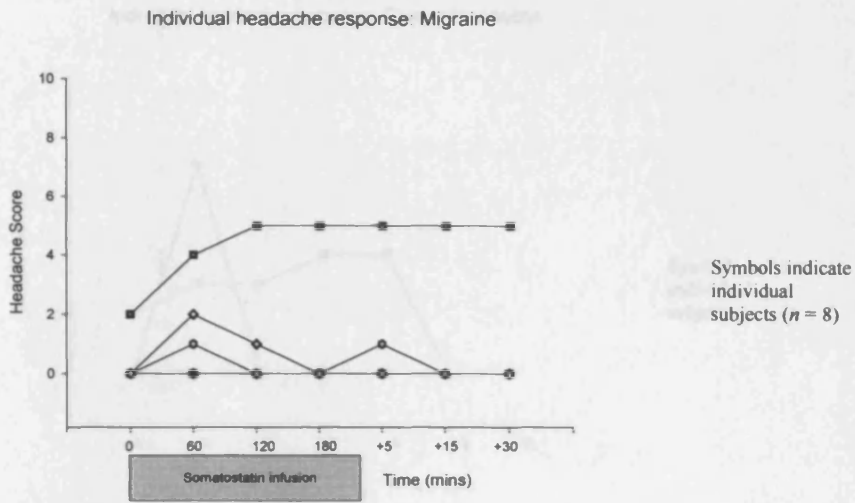
**Figure 20. Effect of somatostatin on GH. Group Data.**



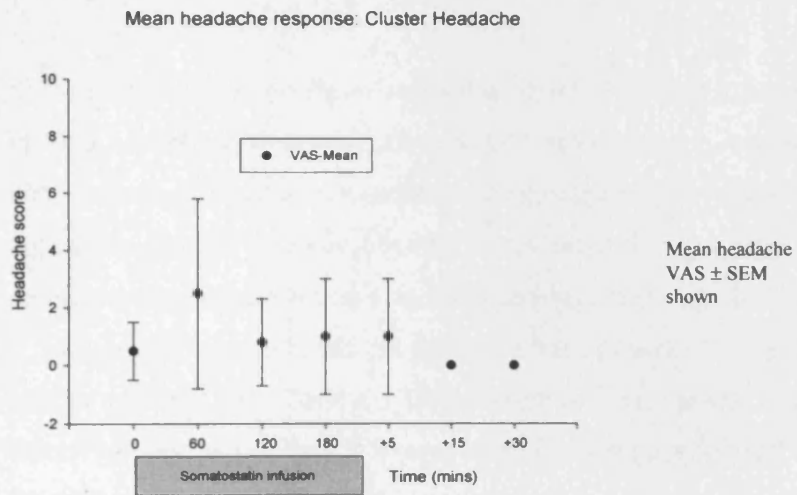
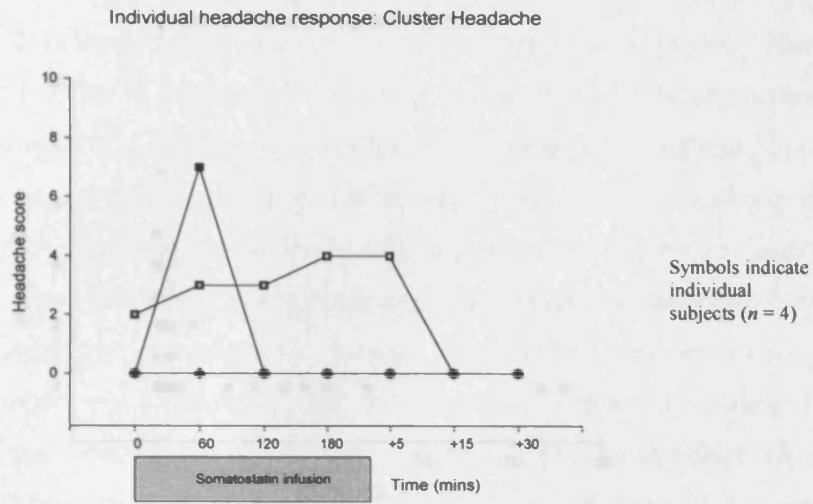
**Figure 21. Effect of somatostatin on headache. Control Group.**



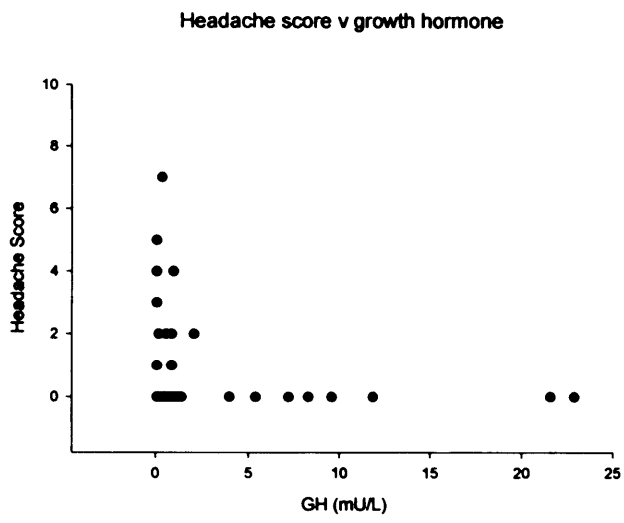
**Figure 22. Effect of somatostatin on headache. Migraine Group.**



**Figure 23. Effect of somatostatin on headache. Cluster Group.**



**Figure 24. Relationship between headache and GH**



### 5.1.6 Discussion

This study demonstrated no significant headache triggering, either immediate or delayed, in patients with migraine or cluster headache, and no headache in control subjects. Significant growth hormone suppression confirmed biologically active somatostatin was infused and an appropriate physiological response obtained. Although one patient in the migraine group and one patient in the cluster headache group had acute pain this response rate is markedly less than that seen with nitroglycerine triggering (Iversen, 2001) and is therefore not considered to be a sufficiently robust response in comparison with nitroglycerin or histamine. Whether that small response represents chance or a biological effect, it is of no value in terms of studying either migraine or cluster headache. The data suggest that somatostatin is not a useful substance in trying to establish a non-vascular migraine-triggering model, although the results do not exclude a role for somatostatin in primary headache.

Somatostatin has a peripheral endocrine effect of lowering first glucagon, and then insulin, as well as its central effect on GH suppression. It is possible that previous observations of headache induction after somatostatin infusion were related to hypoglycemia and peripheral insulin levels rather than a mechanism related to GH secretion. Hypoglycaemia is a recognised trigger for migraine in clinical practice (Lance and Goadsby, 1998) and there has been interest in the relationship between insulin and migraine (Pearce, 1971). Recent evidence points to a genetically determined predisposition to hypoglycemia as a trigger factor for migraine with the identification of single-nucleotide polymorphism alleles in the insulin receptor in some patients with migraine (McCarthy et al., 2001). None of the patients in this study developed hypoglycaemia during the somatostatin infusions.

The observation of octreotide dependent headache suggests that up-regulation of somatostatin receptors is in some way pro-nociceptive (May et al., 1994) and the reciprocal relationship between somatostatin and growth hormone may suggest that GH, or some other signalling molecule related to its secretion, is important in the relationship between somatostatin and pain. Case 3 in the Appendix (Levy et al., 2003b) describes a patient whose headache responded only to octreotide, despite GH reduction with both octreotide and lanreotide, and indicates that the relationship

between somatostatin and pain is not exclusively related to GH. The absence of a correlation between headache score and GH in this study further suggests that somatostatin and GH are not coupled in terms of analgesia.

This study demonstrates that somatostatin infusion-withdrawal alone is not a sufficiently robust stimulus to produce headache. Although I studied a relatively small number of patients the induction rate, particularly in migraine, could not be large enough to be experimentally useful. The role of somatostatin in headache remains to be elucidated, but the anatomical distribution of its receptors strongly supports a role for this peptide in headache. The following chapter investigates the possibility that the acute use of somatostatin analogues may be effective in aborting primary headache.

## **6.Octreotide and primary headache**



## **6.1. Study 5a: Octreotide is not effective in the acute treatment of migraine attacks**

Levy MJ, Matharu MS, Bholra R, Meeran K, Goadsby PJ

Paper ref: Cephalalgia 2004; 24

### **6.1.1 Abstract**

*Objective:* To determine whether subcutaneous octreotide is an effective treatment for acute migraine.

*Methods:* Patients with migraine with and without aura as determined by the International Headache Society were recruited to a double-blind placebo-controlled crossover study. Patients were instructed to treat two attacks of at least moderate pain severity, with at least a seven-day interval, using 100 µg octreotide or matching placebo. The primary endpoint was the headache response defined as: severe or moderate pain becomes mild or nil, at 2 hours. The primary endpoint was analysed using a Multilevel Analysis approach. Secondary end-points included associated symptoms and a four-point functional disability score. The study was powered to detect a 30% difference at an  $\alpha$  of 0.05 and a  $\beta$  of 0.8.

*Results:* A total of 51 patients were recruited, of whom 42 provided efficacy data on attacks treated with octreotide and 41 with placebo. Modelling the treatment outcome as binomial where response was determined by treatment, using the patient as the level 2 variable, and considering a possible period effect, and sex and migraine type as other variables of interest, the effect of subcutaneous octreotide was not significantly superior to placebo. The two hour headache response rates were 20% for placebo and 14% for octreotide, whilst the two hour pain free rates were 7% and 2%, respectively.

*Conclusions:* Subcutaneous octreotide 100 µg is not effective in the acute treatment of migraine when compared to placebo.

### 6.1.2 Introduction

The data in Chapter 4 support the generally accepted view that octreotide has analgesic properties in pituitary tumours. This Chapter investigates the possibility that octreotide may be helpful in primary headache. The aim of Study 5a was to test the hypothesis that octreotide might be effective in the acute treatment of migraine. Although a large number of different triptans, serotonin 5-HT<sub>1B/1D</sub>-agonists, have become available since the discovery of sumatriptan (Ferrari et al., 2001) there is still a need for new classes of abortive agents for migraine. Triptans are contra-indicated in patients with vascular disease, and a group of patients experience universally unacceptable side effects on these agents (Nappi et al., 2003) necessitating the development of novel drugs for migraine. Moreover, many patients overuse 5-HT<sub>1B/1D</sub>-agonists (Silberstein and Liu, 2003) particularly those with long-lasting attacks and headache recurrence, and it is desirable to have more than one pharmacological approach to prevent overuse of a single class of drug.

The involvement of neuropeptides in migraine has been reviewed in Sections 1.11-1.14. The apparent efficacy of CGRP receptor antagonists as abortive agents for migraine (Moreno et al., 2002; Olesen et al., 2004) suggests that the inhibition of nociceptive peptides may be a useful therapeutic approach for migraine. Although substance-P receptor antagonists have proved ineffective in trials of both acute (Goldstein et al., 1997; Diener, 2003; Shephard et al., 1995; Connor et al., 1998; Norman et al., 1998) and preventive (Golstein et al., 2001) migraine management, inhibitory effects have been observed on animal models of trigeminal nociception (O'Shaughnessy and Connor, 1994; Polley et al., 1997). As discussed in Section 1.15, somatostatin is known to have inhibitory effects on a range of neuropeptides, including substance P (Ferrari et al., 1990), CGRP (Helyes et al., 2001) and VIP (Fassler et al., 1990), and it has been proposed that somatostatin may be a potential pharmacological target for the acute treatment of migraine (Selmer et al., 2000). Because native somatostatin is an unstable compound, being broken down by endogenous peptidases within a few minutes (Harris, 1994), octreotide, which has a half-life of approximately 1.5 hours (Harris, 1994), is an attractive compound to study in patients with migraine. In a placebo controlled trial of 29 patients, Kapicioglu et al. (1997) demonstrated that subcutaneous administration of 100 µg octreotide gave

headache relief in 77%, compared to 25% of a parallel placebo-treated group. Despite these findings, no other group has investigated the potential use of somatostatin analogues in migraine. Sicuteri et al. (1984) and Geppetti et al. (1985) found intravenous infusion of somatostatin to be as effective as ergotamine in the acute treatment of cluster headache, although this again has not been systematically further studied and the trial was not sufficiently powered.

As has been reviewed in Section 1.15.2 and Study 4, the analgesic effect of octreotide in acromegaly is well recognised but there are few data regarding its role in primary headache. The onset of octreotide analgesia in acromegaly is more rapid than the 30 minutes required for growth hormone suppression (Chanson et al., 1993), which suggests that octreotide may have a direct anti-nociceptive property. The distribution of sst<sub>2a</sub> within the central nervous system strongly suggests that this particular somatostatin receptor has a role in cranial nociception, being highly expressed in the trigeminal nucleus caudalis and periaqueductal gray (Schindler et al., 1998). Octreotide has particular affinity for the sst<sub>2a</sub> receptor, which may explain its apparent anti-nociceptive action (Schindler et al., 1998).

I sought to verify Kapigioglu et al.'s finding that octreotide is an effective agent for the acute treatment of migraine by studying a larger group of patients in a double-blind placebo controlled cross-over study. I used the more broadly accepted end point of the 2-hour response rate since patients desire quick relief of symptoms (Davies et al., 2000).

### **6.1.3 Subjects and Methods**

Men or women between 18 and 65 years of age were recruited for the study, the majority of patients responding to trial advertisements published in patient information newsletters (The Migraine Trust and Migraine Action Association). All patients interested in the study underwent an initial telephone interview, followed by a detailed face-to-face clinical interview, in order to confirm the diagnosis of migraine with or without aura as defined by the International Headache Society (Headache Classification Committee of the International Headache Society, 1988).

Exclusion criteria included pregnancy and lactation; analgesia or triptan overuse; frequent tension-type headaches (> 10 days/ month); inability to distinguish between tension-type or migraine headache; diabetes mellitus; or a coexisting condition that might expose the patients to a disproportionately increased risk of a significant adverse event: ischaemic heart disease, peripheral vascular disease, cerebrovascular disease, uncontrolled hypertension (blood pressure > 160/95), epilepsy, use of cimetidine, dopamine agonists, cyclosporine or oral hypoglycaemic agents, renal or hepatic impairment. The study was performed in accordance with the ethical principles of the Declaration of Helsinki, and was given local ethical committee approval (Ref 00/N117), as well as Medicines Control Agency (MCA) approval for the administration of octreotide as part of a clinical trial.

### *Design*

The design was a randomised, double-blind, placebo-controlled, single-centre two-attack crossover study, involving patients with acute migraine headache of moderate to severe intensity. Octreotide was kindly donated for the purposes of the trial by Novartis. Octreotide and placebo were packaged into identical pre-filled vials at The Royal Free Hospital pharmacy. All drugs were kept refrigerated in the National Hospital for Neurology and Neurosurgery (NHNN) pharmacy department for the duration of the trial. Normal saline was used as placebo and the dose of octreotide was 100 µg.

At the first visit, patient eligibility for the study was confirmed and a physical examination was performed. Patients who met the inclusion criteria and were willing to participate in the study signed consent agreements authorized by the local ethics committee (NHNN).

Study medication and placebo were provided in two identical pre-filled vials, each containing 1mL of fluid. The vials were labelled Treatment 1 and Treatment 2 for self-administration during a first and second headache attack respectively, each attack being separated by a minimum of 7 days. The order of active treatment and placebo was randomised by NHNN pharmacy, and each participant was assigned a randomisation number. The code was held by the pharmacy until study completion

and database locking. All patients were counselled how to draw up the treatment with a 1mL syringe and taught how to give a self-administered subcutaneous injection. Participants were given the opportunity to practice the injection in supervised conditions during this visit in order to build up confidence. A help line was provided to patients to ring if they experienced trouble during the trial.

### *Efficacy Assessments*

Patients were asked to complete forms during both treatments at home and to report to the investigator (the candidate) within 48 hours of completing both treatments. Headache severity was measured on a 4-point scale (0 = no pain; 1 = mild pain; 2 = moderate pain; 3 = severe pain) at 0 minutes, 30 minutes, 1 hour, 2 hours, 4 hours and 24 hours after study intake. The primary end points for clinical efficacy were the relief of headache, from grade 3 or 2 to grade 1 or 0, within 2 hours after injection for consistency with recent development programmes in acute migraine (Pilgrim, 1991). Additional efficacy measures included headache free rate, functional disability (4 - point scale) and the presence or absence of nausea, vomiting, photo- or phonophobia, all of which were monitored for 24 hours. Rescue medication was permitted 2 hours after the intake of study medication if the headache still required treatment. Ergots and triptans were accepted as rescue medication. Adverse effects were documented on the forms provided after both injections.

### *Statistical Analysis*

The primary efficacy analysis was based on the number of patients who obtained relief from grade 3 or 2 to grade 1 or 0 within 2 hours of treatment. Assuming a treatment difference between placebo and active of 30% based on subcutaneous sumatriptan and placebo response rates (Pilgrim, 1991) it was calculated that 42 patients were needed for the trial to have an 80% power to detect a difference at an  $\alpha$  of 0.05 (Sample Power SPSS Inc.).

The outcome data were to be treated as binary: headache response or none at 2 hours. Because the trial was a cross-over study, attacks 1 and 2 were not truly independent because the patients remained the same, a multivariate analysis approach was

employed using the software that has been developed by the Multi-level Project, MIwiN (available at [www.ioe.ac.uk/multilevel](http://www.ioe.ac.uk/multilevel); Van Vliet et al., 2003).

#### **6.1.4 Results**

A total of 51 patients were recruited, 2 men and 49 women, with a mean age of  $48 \pm 12$  years (mean  $\pm$  SD). Patient demographics are shown in Table 30.

##### *Disposition of patients*

Of the 51 patients recruited, 8 withdrew before completion of the study; one was unable to co-ordinate self-injection during a migraine attack, and seven were lost to follow up; whether they treated or did not treat attacks is not clear. I have treated them as if they did not treat any attacks. One patient was excluded from the first arm of the trial, and two from the second arm because of protocol violation (Figure 25).

##### *Clinical features of study cohort*

Of the 43 patients that successfully completed the study, the frequency of attacks included 4 - 10 per month (35%), 1 - 3 per month (60%) and < 1 per month (5%). The mean duration of attack was  $53 \pm 4.3$  hours. Of the group, 33 (77%) had migraine without aura and 10 (23%) had migraine with aura. Thirty-eight (88%) had previously used 5-HT<sub>1B/1D</sub>-agonists for treatment of acute attacks, of which 28 (74%) experienced meaningful response within 2 hours.

##### *Overall Efficacy*

The primary endpoint of the study was the combined, attack 1 and 2, headache response rate to octreotide at 2 hours compared to placebo. The Wald test was not significant for the overall regression, suggesting that there was no difference between octreotide and placebo even accounting for treatment order, sex, or migraine type.

##### *Efficacy Results*

In total, 42 attacks were treated with octreotide and 41 attacks with placebo. In the octreotide-treated attacks, 6 patients reported headache relief at 2 hours (14%), compared to 8 (20%) in the placebo group.

One patient was rendered pain-free at 2 hours with octreotide (2.4%), whilst 3 patients were rendered pain-free with placebo (7.3%).

The 2-hour- and pain-free response rates are shown in Table 31. Mean pain scores at each time interval are shown in Table 32.

#### *Associated symptoms and functional score*

To evaluate the associated symptoms, only patients who had the symptoms during an attack were included in the analysis. Octreotide performed less well than placebo for nausea, photophobia, phonophobia and aggravation with movement (Figure 26).

There was no difference in functional disability scores between either group at any of the time points post-treatment (Table 32).

#### *Escape Medication*

The frequency of the use of escape medication was no different in the octreotide-treated attacks compared to those treated with placebo: 35 (83%) vs 33 (80%) as shown in Table 33.

#### *Tolerability*

No serious adverse events were reported in either the octreotide- or placebo-treated attacks. Four patients experienced diarrhoea with octreotide, and one patient experienced increased sweating, and another excessive nausea with octreotide, in comparison with their normal attacks. With placebo, one patient reported diarrhoea, one patient experienced light-headedness, and two patients reported excessive tiredness after injection.

**Table 30. Demographic data and migraine characteristics**

---

<b>Characteristic</b>	<b>Patients n = 43 (%)</b>
Age, years (mean $\pm$ SD)	48 $\pm$ 12
Gender n (%)	
<i>Male</i>	2 (5)
<i>Female</i>	41 (95)
Type of migraine n (%)	
MWOA†	33 (77)
MWA††	10 (23)
Frequency of attack per month n (%)	
Daily	0
4-10	15 (35)
1-3	26 (60)
<1	2 (5)
Mean duration of attack $\pm$ SD (hours)	53 $\pm$ 4.3
Previous 5-HT <sub>1B/1D</sub> -agonist use	38 (88)
Meaningful response to 5-HT <sub>1B/1D</sub> -agonists	28 (74)

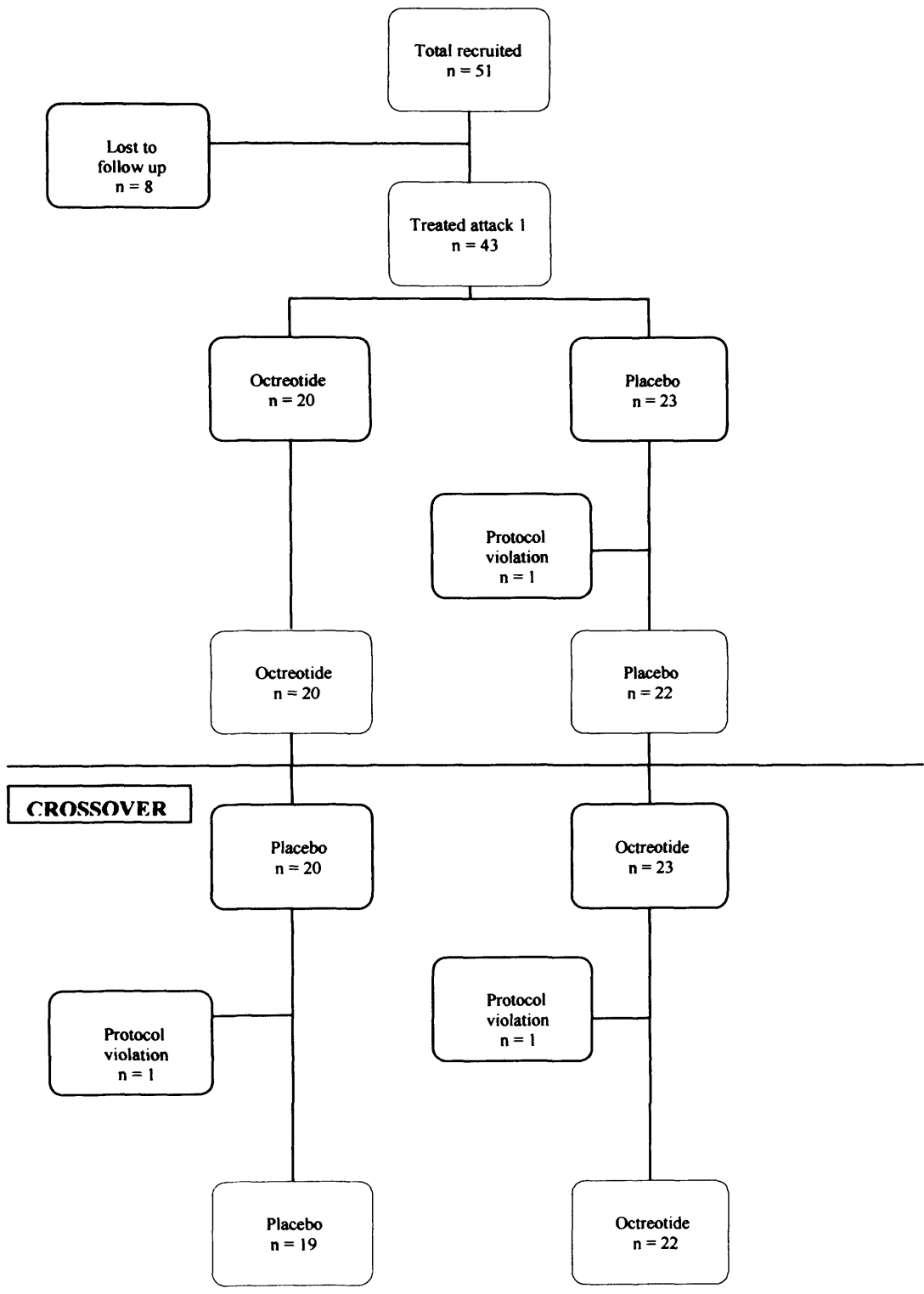
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† MWOA : migraine without aura

†† MWA : migraine with aura



**Figure 25. Disposition of patients in study**



**Table 31. Two hour- and pain free- response rates**

<b>Groups</b>	<b>Not improved at 2 hours (%)</b>	<b>Headache Response at 2 hours† (%)</b>	<b>Pain-free at 2 hours†† (%)</b>	<b>Total no. treatments</b>
Placebo	33 (81)	8 (20)	3 (7)	41 (49)
Octreotide	36 (86)	6 (14)	1 (2)	42 (51)
<b>Total</b>	<b>69 (83)</b>	<b>14 (17)</b>	<b>4 (5)</b>	<b>83 (100)</b>

† Headache response: relief of headache from moderate / severe/ excruciating pain to nil or mild pain

†† Pain free: relief of headache from moderate / severe/ excruciating pain to no pain

**Table 32. Mean pain and functional disability scores (0-3)**

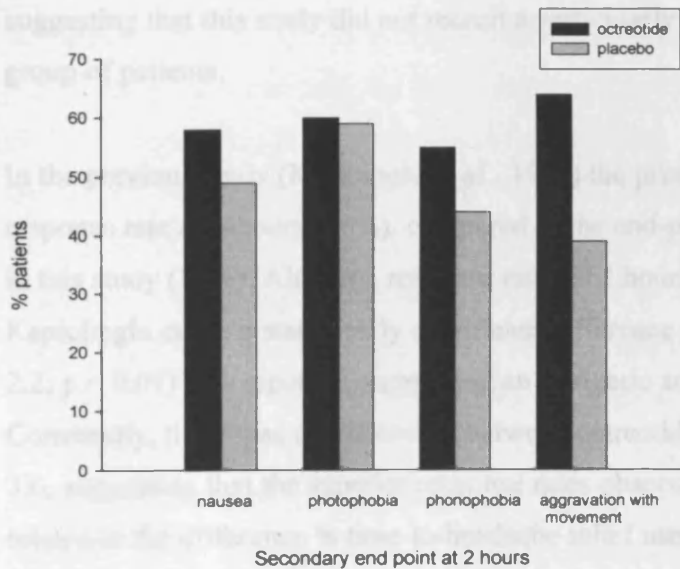
	<b>Group</b>	<b>0 minutes</b>	<b>120 minutes</b>	<b>240 minutes</b>	<b>24 hours</b>
<b>Pain Score</b>	<b>Placebo</b>	2.3 ± 0.5	2.2 ± 1.0	2.0 ± 1.0	1.3 ± 1.2
	<b>Octreotide</b>	2.3 ± 0.6	2.2 ± 0.8	2.0 ± 1.0	1.2 ± 1.1
<b>Functional Score</b>	<b>Placebo</b>	1.6 ± 1.0	1.9 ± 1.1	1.7 ± 1.2	1.0 ± 1.1
	<b>Octreotide</b>	1.5 ± 0.9	1.9 ± 1.0	1.7 ± 1.0	1.4 ± 1.2

4.1.5 Discussion

**Table 33. Escape medication use**

	Octreotide n (%)	Placebo n (%)
Escape medication used	35 (83)	33 (80)
Escape medication not used	7 (17)	8 (20)
<b>Total</b>	<b>42 (100)</b>	<b>41 (100)</b>

**Figure 26: Percentage of patients reporting persistent associated symptoms at 2 hours**



### 6.1.5 Discussion

Contrary to previous reports, the results of this study do not show a difference between 100µg octreotide and placebo in the acute treatment of migraine. I did not find octreotide to be superior to placebo for relief of headache, associated symptoms or functional disability scores.

It is not clear why these findings are so different to the previous study (Kapicioglu et al., 1997). The placebo response rate of 20% is not different from the 25% placebo in the previous study (Kapicioglu et al., 1997) suggesting that there was little difference in the patient population group in terms of migraine characteristics. However, the relatively low placebo response rates in both studies suggest that there may have been a selection bias towards a group of patients with difficult migraine (Jhee et al., 1998). This seems likely given that patients who volunteered were willing to have an injection for their migraine attacks. The mean attack duration in the study group was 2.2 days, which is longer than that seen in population studies (Koseoglu et al., 2003). Despite this, 74% of patients reported a useful response with 5-HT<sub>1B/1D</sub>-agonists, suggesting that this study did not recruit a particularly pharmacologically refractory group of patients.

In the previous study (Kapicioglu et al., 1997) the primary efficacy end-point was response rate at 6-hours (76%), compared to the end-point of response rate at 2 hours in this study (14%). Although response rates at 2 hours were not provided by Kapicioglu et al., a statistically significant difference in pain scores at 2 hours (1.5 v 2.2;  $p < 0.01$ ) was reported, suggesting an analgesic action at this earlier time point. Conversely, there was no difference between octreotide and placebo at 6 hours (Table 33), suggesting that the superior response rates observed in the earlier study were not related to the difference in time-to-headache relief used in the two studies. It is generally considered that if an acute treatment for migraine is to be useful in clinical practice, it should have an effect within 2 hours (Geraud et al., 2000). For an injectable treatment this is considered slow, so my primary end point could be considered generous.

The study by Kapigioglu et al. (1997) did not have a cross-over design, so it is possible that the excellent response rate they found in the treatment group was due to a difference in migraine characteristics from the placebo-treated group. My study had a cross-over design to avoid this possible confounder. The previous study (Kapicioglu et al., 1997) did not give outcome data for the secondary end-points of associated symptoms or functional ability scores, although my findings were negative. The gastro-intestinal side effects observed in five patients in response to 100 µg octreotide in our study is in keeping with the reported experience of side effects on this dose (Plockinger et al., 1990). The same dose of octreotide was used as in the previous study (Kapicioglu et al., 1997) suggesting that the difference in observed response is not dose-related.

Although the findings were negative, the somatostatin receptor remains a theoretically attractive pharmacological approach to primary headache management. As discussed in Section 1.15.1, there are *in vivo* animal data to suggest that the central administration of somatostatin and its analogues results in anti-nociception, both in peripheral (Helmchen et al., 1995) and trigeminal (Bereiter, 1997) pain models. In conjunction with the localisation of somatostatin receptors (particularly sst<sub>2</sub>) in the trigeminal nucleus caudalis and periaqueductal gray (Schindler et al., 1996) these findings suggest that drugs acting on the somatostatin receptor remain realistic therapeutic options. It is possible that octreotide needs to cross the blood-brain barrier to have an effect in primary headache, whilst it may not need to do this to be analgesic in pituitary tumour-associated headache. If this is the case, octreotide may simply be the wrong compound due to its non-lipophilicity and apparent poor ability to cross the blood-brain barrier (Kitazawa et al., 1998; Schmidt et al., 1998). It is necessary to develop a somatostatin analogue with good brain access to test the somatostatin theory completely. Novel somatostatin analogues with differing receptor affinities are currently being developed for the treatment of functional pituitary adenomas (Shimon et al., 1997). Some of these compounds are particularly avid for the sst<sub>2</sub> receptor, making them potential targets for analgesic agents (Saveanu et al., 2002). The central nervous system permeability of these new agents has not yet been determined, although they present attractive compounds to study for future clinical and non-clinical trials as regards primary headache and requires further study.

## **6.2. Study 5b: subcutaneous octreotide is effective in the treatment of acute cluster headache**

Matharu MS, Levy MJ, Meeran K, Goadsby PJ.

Abstract ref: Cephalalgia (2003) 23: 687. 6.4

Paper ref: Annals of Neurology 2004 (in press)

### **6.2.1 Abstract**

*Objective:* To determine whether subcutaneous octreotide is an effective treatment for acute cluster headache.

*Methods:* Patients with episodic and chronic cluster headache, as defined by the International Headache Society, were recruited to a double-blind placebo-controlled crossover study. Patients were instructed to treat two attacks of at least moderate pain severity, with at least a 24 hour break, using subcutaneous octreotide 100µg or matching placebo. The primary endpoint was the headache response defined as: very severe, severe or moderate pain becomes mild or nil, at 30 min. The primary endpoint was analysed using a Multilevel Analysis approach.

*Results:* A total of 57 patients were recruited of whom 46 provided efficacy data on attacks treated with octreotide and 45 with placebo. The headache response rate with subcutaneous octreotide was 52 % while that with placebo was 36 %. Modelling the treatment outcome as a binomial where response was determined by treatment, using the patient as the level 2 variable, and considering period effect, sex and cluster headache type as other variables of interest, the effect of subcutaneous octreotide 100µg was significantly superior to placebo ( $P < 0.05$ ).

*Conclusions:* Subcutaneous octreotide 100 µg is effective in the acute treatment of cluster headache when compared to placebo.

## 6.2.2 Introduction

The previous study (5a) found octreotide to be ineffective in the acute treatment of migraine. The current study investigates the possibility that octreotide may be helpful for the acute treatment of cluster headache (CH). As described in Section 1.5.1, CH is the most severe form of primary neurovascular headache. It is characterized by excruciating pain lasting 15 to 180 minutes (Headache Classification Committee of The International Headache Society, 2004). Controlled evidence exists to treat acute attacks of CH with oxygen inhalation (Fogan, 1985), intranasal (van Vliet et al., 2003) and injectable (Ekbom, 1991) sumatriptan, high dose oral zolmitriptan (Bahra et al., 2000) and intranasal dihydroergotamine (Andersson & Jespersen, 1986). An unequivocally non-vasoconstrictor treatment for acute CH is not available. Ergots and triptans are contraindicated in patients with vascular disease and therefore caution must be exercised in patients with CH since the disorder predominates in middle-aged men, who often have risk factors for cardiovascular disease, particularly smoking (Manzoni et al., 1983). There is, therefore, a compelling need to develop new pharmacological approaches to CH, particularly if possible approaches without vascular effects, in order to effectively and safely manage these patients. Moreover, the question of whether a non-vasoconstrictor approach might be effective would offer a fundamental insight into the more generic issue of whether CH is a vascular or central nervous system disorder.

As described in Sections 1.11 and 1.14, an acute cluster attack is associated with the release of calcitonin gene related peptide (CGRP) and vasoactive intestinal polypeptide (VIP), while triptans, serotonin 5-HT<sub>1B/1D</sub> agonists, attenuate the levels of these neuropeptides during successfully treated attacks, thereby implying that the inhibition of these neuropeptides is an important mechanism for aborting a cluster attack (Goadsby & Edvinsson, 1994; Fanciullacci et al., 1995). Therefore, octreotide is an attractive compound to investigate, for the same reasons as described in the previous migraine study.

Only two previous studies have been performed which have looked at the potential benefits of octreotide in CH. In the first study, intravenous somatostatin (25 µg/min for 20 minutes) was compared to treatment with ergotamine (250 µg intramuscularly),

or placebo in a double-blind trial comprising 72 attacks in 8 patients (Sicuteri et al., 1984). Infusion of somatostatin reduced the maximal pain intensity and the duration of pain significantly compared to placebo, and to a degree comparable to intramuscular ergotamine. In another randomized, double-blind study subcutaneous somatostatin was compared with ergotamine (Geppetti et al., 1985). Five patients were treated for three attacks by each of the drugs. Subcutaneous somatostatin and ergotamine were equally beneficial as regards effects on maximal pain intensity and the pain area, but somatostatin was less effective in reducing the duration of pain. This limited evidence of the beneficial effect of somatostatin needs to be explored further in properly controlled and adequately powered studies.

The aim of the current study was determine whether octreotide is an effective abortive agent for the acute treatment of CH.

### **6.2.3 Subjects and Methods**

#### *Patients*

Men or women between 18 and 65 years of age with an established diagnosis of CH, according to the International Headache Society (Headache Classification Committee of the International Headache Society, 2004), were recruited for this single centre study. Patients were required to have CH attacks of at least 45 minutes duration when untreated. The ethical considerations and exclusion criteria are the same as for the migraine study (Study 5a).

#### *Design*

This was a randomised, double-blind, two-attack, crossover study of 100 µg subcutaneous octreotide and matching placebo. The packaging and storage of octreotide and placebo, and patient education regarding self-injection was identical to the migraine study. Patients were asked to treat two attacks at least 24 hours apart with either octreotide or matching placebo in a randomised order. They were instructed to grade attacks on an ordinal categorical (five-point) scale of none, mild, moderate, severe or very severe (Pilgrim, 1991). Subsequent assessments were at 5, 10, 15, 20, 30 and 60 minutes. Escape medication was allowed at 30 minutes post-



dose, usually injectable or intranasal sumatriptan, oxygen or analgesic, but not an ergotamine derivative.

### *Efficacy Assessments*

The primary outcome measure was headache severity at 30 minutes, a reduction in headache from moderate, severe, or very severe to nil or mild. Secondary outcome measures included the percentage of patients headache-free at 30 minutes, rate of relief of associated symptoms, time to initial relief and rate of meaningful relief. Associated symptoms, such as vomiting, nausea, photophobia, phonophobia, lacrimation, nasal congestion, and other autonomic features, were recorded immediately before treatment and at 30 minutes. Initial relief was defined as the time that a patient recorded any headache relief. Patients were asked if they considered the response at 30 minutes meaningful.

### *Statistical Analysis*

Using the results of a crossover study of subcutaneous sumatriptan versus placebo, and assuming a treatment difference between placebo and active of 30%, it was calculated (using Sample Power) that 42 patients were needed for the study to have an 80% power to detect a difference at an  $\alpha$  of 5%. The outcome data were treated as binary: headache response or none at 30 minutes. As for study 5a, the effect of active treatment and attack order was examined, as well as other variables of interest, such as sex, site, and CH type. Considering that attacks 1 and 2 are not strictly independent because the patients remain the same, a multilevel multivariate analysis was employed using the software that has been developed by the Multilevel Project, MLwiN (available at [www.ioe.ac.uk/multilevel](http://www.ioe.ac.uk/multilevel); Van Vliet et al., 2003).

#### 6.2.4 Results

A total of 57 patients were recruited, 45 men and 12 women, with a mean age of  $40 \pm 10$  years (mean  $\pm$  SD).

##### *Disposition of patients*

Of the 57 patients recruited, six came to the end of the bout before completing the study; one of these treated the first attack. Two patients withdrew before treating any attacks, and two were lost to follow up; whether they treated or did not treat attacks was not clear, and they have been treated as if they did not treat any attacks. Two attacks of mild severity were treated, and prior to the treatment of two attacks the syringe malfunctioned hence the patients were unable to treat the attacks; these four attacks were excluded from the analysis. Use of escape medication before 30 minutes after treatment was reported in five attacks, which were scored as outcome failures (Figure 27).

##### *Clinical features of study cohort*

The mean duration of CH history was  $14 \pm 9$  years (mean  $\pm$  SD). Forty-one patients had episodic CH, fifteen had chronic CH, and one was unclassifiable because it was the first bout. The average bout length of the patients with episodic CH was  $9 \pm 5$  weeks (mean  $\pm$  SD). The average attack duration at recruitment was reported by patients to be  $107 \pm 75$  minutes (mean  $\pm$  SD). Thirty-seven patients had previously used sumatriptan injection, of whom 36 were responsive; 25 had used intranasal sumatriptan, of whom 16 were responsive; 38 had used oral sumatriptan, of whom 12 were responsive. In terms of previous use of oxygen, 15 had used high dose and high flow oxygen, of whom 9 were responsive; 9 used low dose or low flow rate oxygen, of whom 4 were responsive.

##### *Overall efficacy*

The primary endpoint of the study was the combined, attack 1 and 2, headache response rate at 30 minutes compared to placebo. The Wald test was significant for the overall regression ( $\chi^2 = 14.1$ ,  $P = 0.007$ ) with only the treatment term being

significantly different from zero. There was no significant effect of treatment order, cluster headache type and gender.

### *Efficacy Results*

In total, 46 attacks were treated with octreotide and 45 with placebo. In the octreotide-treated attacks, 24 patients reported headache relief at 30 minutes (52 %) compared to 16 (36 %) patients who treated an attack with placebo (Table 35). Fifteen patients were pain-free at 30 minutes (33 %) when treated with octreotide, compared to 6 (13 %) when the attack was treated with placebo ( $\chi^2 = 9.8$ ,  $P = 0.04$ ; Figure 28).

### *Associated symptoms*

To evaluate the associated symptoms, only patients who had the symptom immediately before treatment were included in the analysis. Conjunctival injection, lacrimation, nasal congestion, rhinorrhoea, ptosis, eyelid oedema and photophobia were the most frequently mentioned. In the attacks treated with octreotide, more patients experienced relief from associated symptoms at 30 minutes (Figure 29).

### *Time to Initial Relief*

The mean time to initial relief in the octreotide-treated group was  $18.3 \pm 8.9$  minutes, compared to  $18.1 \pm 7.0$  minutes in the placebo-treated group.

### *Meaningful relief*

Patients were asked if they thought the CH attack was adequately treated at 30 minutes. Seventeen (37 %) patients who treated an attack with octreotide reported meaningful relief, compared to 13 (29 %) patients who had treated an attack with placebo.

### *Escape Medication*

The frequency of use of escape medication was lower in octreotide-treated attacks compared to those treated with placebo; 20 (44%) vs 25 (56%).

### *Tolerability*

No serious adverse effects were reported with either the octreotide- or placebo-treated attacks. Eight patients treated with octreotide (17 %) reported minor gastrointestinal disturbance, including nausea, abdominal bloating and diarrhoea, compared with 4 patients (9 %) treated with placebo. The adverse events are shown in Table 36. All resolved spontaneously and were generally short-lived and mild in nature.

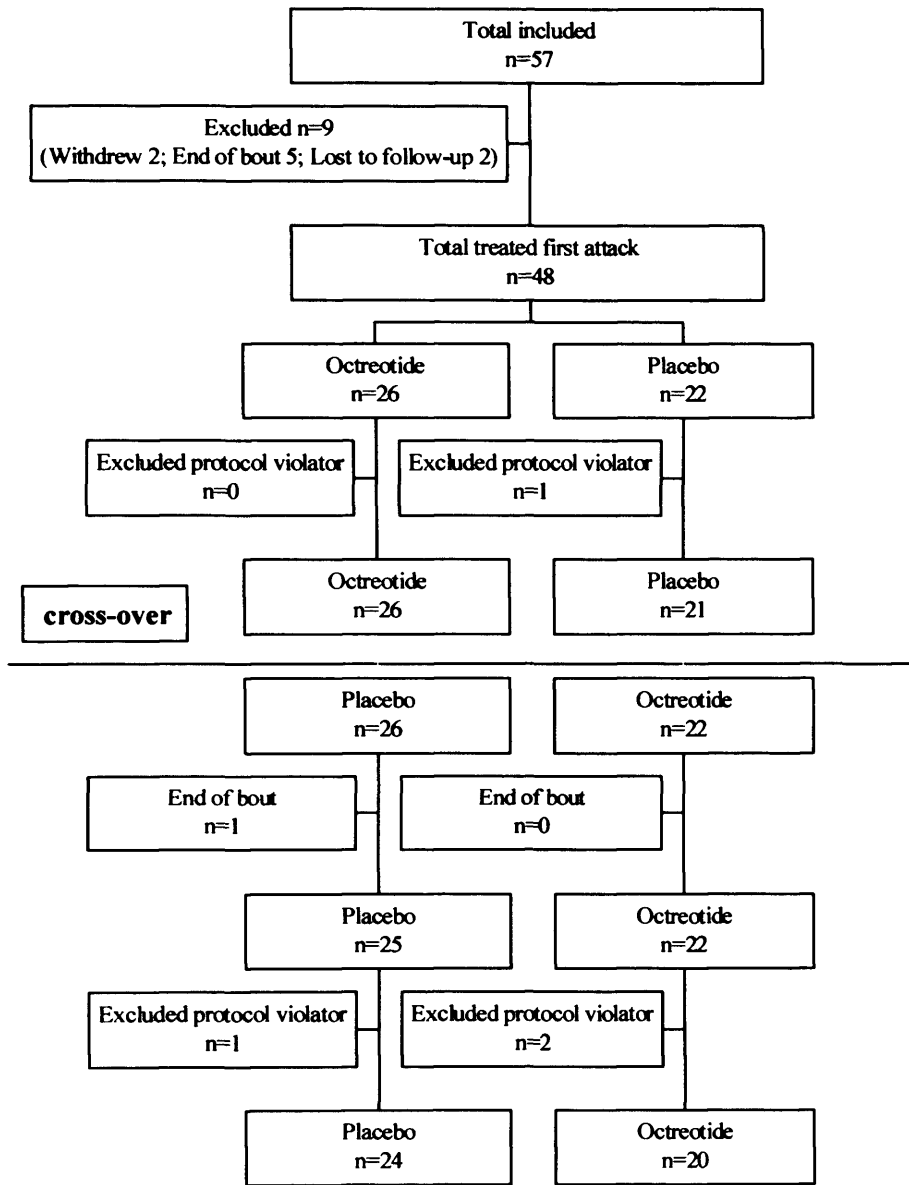
**Table 34. Demographic data and cluster headache characteristics**

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Characteristic	Patients n = 57 (%)	
<i>Age, years (mean ± SD)</i>	40	± 10
<i>Gender, n (%)</i>		
Male	45	(79)
Female	12	(21)
<i>Type of CH, n (%)</i>		
Episodic	41	(72)
Chronic	15	(26)
Unclassifiable	1	(2)
<i>Mean attack duration ± SD (minutes)</i>	107	± 75
45 – 60	21	(37)
61 – 90	14	(25)
91 – 180	16	(28)
> 180	6	(11)
<i>Mean bout duration ± SD (weeks)</i>	9	± 5
<i>Length of history ± SD (years)</i>	14	± 9

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**Figure 27. Disposition of patients in study**



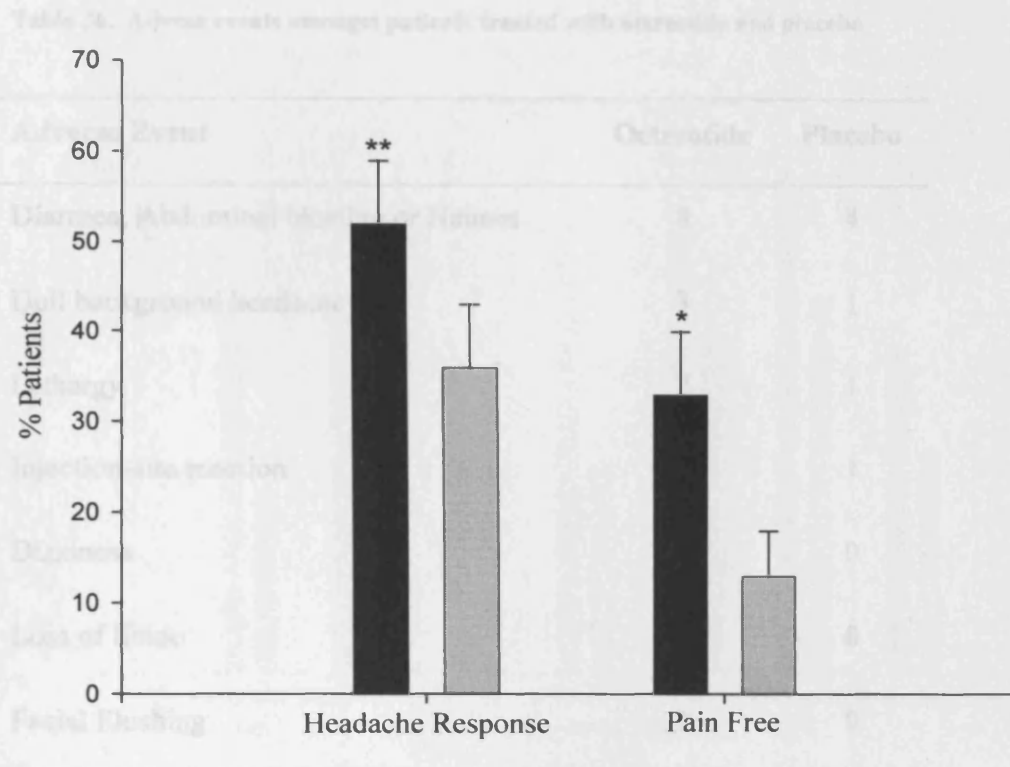
**Table 35. Efficacy of octreotide and placebo**

<b>Groups</b>	<b>Not improved at 30 minutes (%)</b>	<b>Headache Response† at 30 minutes (%)</b>	<b>Pain-free†† at 30 minutes (%)</b>	<b>Total no. treatments</b>
Placebo	29 (64)	16 (36)	6 (13)	45 (49)
Octreotide	22 (48)	24 (52)	15 (33)	46 (51)
Total	51 (56)	40 (44)	21 (23)	91 (100)

† Headache response: relief of headache from moderate / severe/ excruciating pain to nil or mild pain

†† Pain free: relief of headache from moderate / severe/ excruciating pain to no pain

Figure 28. Efficacy of octreotide versus placebo in cluster headache



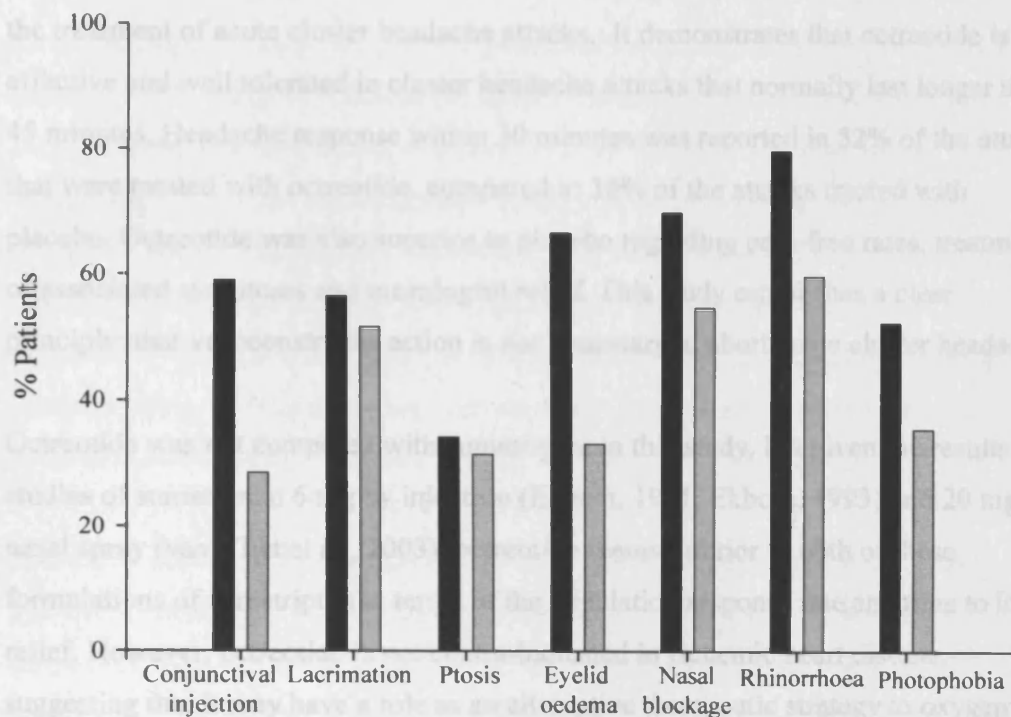
Efficacy: headache response (a reduction of headache intensity from very severe, severe or moderate to mild or no pain) and pain-free rates (no pain) at 30 minutes after treatment with octreotide (■) versus placebo (■). \*\*  $P < 0.01$ ; \*  $P < 0.05$ .



**Table 36. Adverse events amongst patients treated with octreotide and placebo**

<b>Adverse Event</b>	<b>Octreotide</b>	<b>Placebo</b>
Diarrhea, Abdominal bloating or Nausea	8	4
Dull background headache	3	1
Lethargy	2	1
Injection-site reaction	2	1
Dizziness	1	0
Loss of libido	1	0
Facial Flushing	1	0

**Figure 29. Percentage improvement in associated symptoms at 30 minutes**



■ Octreotide

■ Placebo

\* Only patients with symptoms at baseline were included. Because of multiple-comparison issues, no statistical analysis was performed.

### 6.2.5 Discussion

This is the first placebo-controlled trial investigating the potential use of octreotide in the treatment of acute cluster headache attacks. It demonstrates that octreotide is effective and well tolerated in cluster headache attacks that normally last longer than 45 minutes. Headache response within 30 minutes was reported in 52% of the attacks that were treated with octreotide, compared to 36% of the attacks treated with placebo. Octreotide was also superior to placebo regarding pain-free rates, treatment of associated symptoms and meaningful relief. This study establishes a clear principle: that vasoconstrictor action is not necessary to abort acute cluster headache.

Octreotide was not compared with sumatriptan in this study, but given the results of studies of sumatriptan 6 mg by injection (Ekbom, 1991; Ekbom, 1993) and 20 mg by nasal spray (van Vliet et al., 2003), octreotide seems inferior to both of these formulations of sumatriptan in terms of the population response rate and time to initial relief. However, octreotide is not contra-indicated in ischemic heart disease, suggesting that it may have a role as an alternative therapeutic strategy to oxygen in patients who cannot take 5-HT<sub>1B/1D</sub> receptor agonists. It is important to recall that the effect in responders is no less than the effect of injectable sumatriptan, since the response endpoint is identical, but that the study suggests the population of responders is smaller.

Octreotide was well-tolerated with no reports of serious side effects. The main side effect observed was gastrointestinal upset in eight patients treated with octreotide compared to four patients treated with placebo. All side effects resolved spontaneously and were generally short-lived and mild in nature. The side effects reported in this study were in keeping with the reported experience of side effects with this drug (Newman et al., 1995). The good tolerability data makes octreotide an attractive agent in a condition in which repeated doses will be required over a relatively short. Long-term octreotide therapy is commonly used in patients with acromegaly, and although there is a theoretical risk of gall stone development, this rarely poses a significant clinical problem compared to the burden of the disease itself (Lancranjan & Atkinson, 1999). Hence, the prolonged use of octreotide in CH, with

its concomitant devastating morbidity from the excruciating pain, appears to be reasonable on clinical grounds.

As in the migraine study, the crossover approach was used. This design is particularly attractive in the study of CH because attacks are relatively stereotyped, and the attacks occur in rapid succession (van Vliet et al., 2003). Because treatment of two separate attacks in the same patient means that the attacks are not strictly independent, a multilevel multivariate statistical approach was used. The analysis showed no influence of the order in which the treatment was given. Moreover, the use of the multilevel multivariate approach permits consideration of all the relevant data in a single analysis. This analysis has been used in a randomized controlled trial of sumatriptan nasal spray in cluster headache (Van Vliet et al., 2003).

There are some limitations of this study that require consideration. First, patients were warned about about potential gastrointestinal side effects from octreotide when consenting patients for the study, in line with the requirements of the local ethics committee. This may have introduced an element of unblinding to the study in the patients who experienced these symptoms, which may have biased the results. This did not manifest as a significant ordering effect in the analysis. Secondly, this was a single dose study, and long-term studies would need to be performed to determine the long-term use of this medication, since cluster headache is a chronic condition. Thirdly, the study population consisted of 72% episodic cluster patients and 26% chronic patients. Although no difference in response was observed between the two, the study was not designed to dissect out the more common episodic from chronic cluster headache. A separate study of chronic cluster headache may reveal less robust results, which would, on the other hand, predict a better outcome in episodic cluster headache should a larger study in that sub-group be done. Lastly, this is a single centre study, although the unit in which this study was performed (QS) acts as a referral centre for the entire UK National Health Service, so the patient mix is taken from right across the UK.

As has been discussed in Section 1.5.1, functional imaging with positron emission tomography (PET; May et al., 1998a), structural imaging with voxel-based

morphometry (May et al., 1999) and exciting results with deep brain stimulation (Leone et al., 2001) have identified the posterior hypothalamic gray matter as the key area for the basic defect in CH. Given that the hypothalamus is the chief source of somatostatin within the central nervous system (Swaab et al., 1993), the alteration in hypothalamic activity in CH may result in the disinhibition of descending somatostatinergic pathways to the trigeminal nucleus caudalis, either directly or via the periaqueductal gray. There is evidence that the periaqueductal gray and trigeminal nucleus caudalis are under tonic inhibition from the hypothalamus (Lumb, 2002). The disinhibition of descending input caused by hypothalamic dysfunction could theoretically result in uncontrolled trigeminovascular activation, as seen during a cluster headache attack. This hypothetical state of reduced somatostatinergic inhibitory activity may explain the observation of reduced somatostatin levels during CH (Caleri et al., 1987), as well as the therapeutic effect of administering exogenous somatostatin (Sicuteri et al., 1984; Geppetti et al., 1985) and octreotide, as found in this study.

Finally, these results are interesting in the context of the negative migraine study. The striking difference between migraine and cluster headache in the pattern of brain activation on functional imaging, where cluster headache patients activate the posterior hypothalamus (May et al., 1998a; Sprenger et al., 2004), and both episodic (Weiller et al., 1995; Bahra et al., 2001) and chronic (Matharu et al., 2003) migraine the brainstem without hypothalamic activation, is in keeping with greater prominence of a somatostatinergic mechanisms in cluster headache. If this were so, this study would be the first substantial evidence and pharmacologically based difference in the acute treatment of these disorders, which are so strikingly clinically different.

As discussed in Section 1.15.1, somatostatin has been found to differentially modify the release of a variety of neurotransmitters in several regions of the brain. Serotonin release from rat hypothalamic, cortical and hippocampal slices is enhanced by somatostatin (Tanaka & Tsujimoto, 1981), as is noradrenaline release from the cortex (Tsujimoto & Tanaka, 1981), whilst noradrenaline release is inhibited in the rat hypothalamus (Gothert, 1980) and chick sympathetic ganglia (Boehm & Huck, 1996), and the release of  $\gamma$ -amino butyric acid in the rat striatum is inhibited (Meyer et al.,

1989). Further studies will be required to understand the mechanism of action of somatostatin and its analogues in CH.

In conclusion, this is the first adequately powered placebo-controlled study to demonstrate the effectiveness of a somatostatin analogue in the treatment of acute cluster headache attacks. In clinical practice, octreotide may have a particular utility in patients who are unresponsive to or intolerant of 5HT<sub>1B/1D</sub> agonists and oxygen, and as an alternative to oxygen in patients with cardiovascular disease. Furthermore, this study demonstrates that somatostatin analogues which have no vasoconstrictor effect offer a novel therapeutic approach to the treatment of acute cluster headaches that may offer insights into understanding more fundamental aspects of this disabling form of primary headache.

## **7. General Discussion**

## **7.1 General Discussion**

This thesis has investigated the role of the hypothalamo-pituitary axis in headache. The content of this thesis essentially consisted of two components. The first component investigated the structural, biochemical and clinical aspects of pituitary tumour-associated headache. The second component investigated the role of somatostatinergic pathways in primary headache.

The negative findings of Study 1 cast doubt on the accepted view that pituitary tumour-associated headache is a purely structural problem and suggest that other factors must be involved in its pathophysiology. This is an important observation because headache is widely regarded amongst many physicians to be a direct result of either dural stretch or irritation of pain-producing structures within the cavernous sinus. Although the relevance of intrasellar pressure was not addressed in this thesis, the question of whether pituitary tumour-associated headache has a biochemical component is further emphasised.

Study 2 attempted to isolate potentially nociceptive peptides within pituitary tumours. Although both CGRP and substance P were isolated within a variety of tumour subtypes, there was no robust association with headache. Other candidate peptides presented in the general introduction include neuropeptide Y (NPY) and vasoactive intestinal polypeptide (VIP), and the possibility remains that these, or other peptides, may be involved in pituitary tumour-associated headache. However, although not presented in this thesis, preliminary data from our group also suggests that there is also no robust relationship between NPY (Classey et al., 2003) or VIP (Nathoo, unpublished data) with headache. This either raises the question of the validity of the nociceptive peptide hypothesis, or suggests that the relationship between pituitary tumours and headache is more complex than the production of a single nociceptive peptide.

Throughout this thesis, growth hormone- and prolactin- secreting tumours were particularly associated with headache. In Study 1, these tumour types were associated with the highest headache scores, and in Study 3, they were associated with the



highest disability from headache. Although the precise mechanisms of nociception in these tumour types remain elusive, it strongly suggests that biochemical factors are important in the headache associated with these tumour types. This argument is reinforced by the observation in Study 3 that rare headache types such as SUNCT were exclusively associated with acromegaly and prolactinomas, and the finding that dopamine agonists and somatostatin analogues were able to alter the headache characteristics in these tumour types.

Pituitary tumour-associated headache is not a single phenotype but is a heterogeneous clinical spectrum of presentation, which appears to be governed both by tumour- and patient-specific factors. Migraine was the commonest headache presentation, occurring in approximately 80% of cases. That migraine is the commonest headache type in pituitary tumours has since been confirmed by the findings of other groups interested in this topic (Afra & Czirjak, 2003). It is unclear whether the occurrence of migraine in the presence of a pituitary tumour is due to expression of migraine in predisposed individuals, or whether the tumour caused the migraine *de novo* in certain cases. The finding of trigeminal autonomic cephalgias in a relatively small cohort of patients in Study 3 further highlights the possibility that hypothalamo-pituitary disturbance is important in these headache syndromes. It was observed that patients with pituitary tumours may continue to suffer with debilitating headache after surgery, and that phenotype-driven medical therapy is helpful in many cases. It was observed that patients with ipsilateral cavernous sinus invasion were particularly refractory to medical therapy, and there appears to be a sub-set of patients with unclassifiable headache that may be specific to pituitary disease. The observations of Study 3 have led to a proposed modification of the current classification of pituitary tumour-associated headache.

The theoretical reasons why somatostatin might play an important role in headache have been discussed in this thesis. The interesting difference in response between cluster headache patients and migraineurs to octreotide may raise important lessons regarding the pathophysiology of these two disorders. There appeared to be a real effect of octreotide on cluster headache, and further studies are required to determine whether this pharmacological approach may be useful in clinical practice. As regards octreotide and pituitary tumour-associated headache, Study 3 confirmed the

impression that octreotide is often analgesic in pituitary tumours. This study also highlighted the less well recognised fact that octreotide overuse can lead to worsening headache in many situations, which provides a cautionary tale in the use of this agent for headache. In the case of somatostatin overexposure and headache triggering, it does not appear that this will be a useful tool for experimental headache induction.

## 7.2 Conclusion and further direction

The scientific study of headache is in its relative infancy compared to other areas of medical research, and it is an exciting time to be involved in the field. There is much to be learned regarding the role of the hypothalamo-pituitary axis in headache, and the findings in this thesis have contributed to this area. In recent years, there has been a large expansion in the number of publications regarding functional imaging and headache, *in vivo* models of trigeminal nociception, and clinical studies regarding the presentation and management of headache. The *in vivo* models allow for a great opportunity to further study the neuroendocrine mechanisms involved in headache. For example the effect of micro-injecting certain peptides into the hypothalamus on trigeminal activity may represent an exciting opportunity to further understand specific neuroendocrine pathways in headache. Although not presented in this thesis, our group has studied the effect of micro-injecting various compounds into the posterior hypothalamic nucleus, and have found these to have profound effects on trigeminal nerve firing. For example, we have shown that somatostatin antagonists inhibit trigeminal activity (Bartsch, unpublished data) and have also shown that injection of orexin A inhibits, whilst orexin B significantly facilitates trigeminal activity (Bartsch et al., 2004). This opens up a relatively new area of research opportunities for both neurologists and neuroendocrinologists interested in headache.

In conclusion, the subject of headache has traditionally been within the realms of the neurologist and neuroscientist. The finding that disruption of the hypothalamo-pituitary axis can lead to interesting headache syndromes suggests that endocrinologists may contribute significantly to this rapidly expanding field of medicine.

## 8. Appendix

### 8.1 Case 1

#### *Micro-prolactinoma and SUNCT-like headache*

Levy MJ, Matharu MS, Goadsby PJ

Paper ref: European Journal of Neurology 2003;10:1-5

A 36-year-old female, with no headache history, presented in 1987 with secondary amenorrhoea, galactorrhoea and headache. The headache was described as a left-sided sharp 'knife-like' pain during acute exacerbations. An exacerbation would typically last between 10 seconds and 4 minutes and would occur up to 20 times a day. Precipitants for exacerbations included bending, walking and moving about. In between these exacerbations, she had a continuous dull left-sided pain. During an exacerbation, she experienced prominent ipsilateral cranial autonomic symptoms of lacrimation, nasal stuffiness and ptosis. She felt irritable during the pain and would prefer to move around rather than lie still. There was no associated nausea, photophobia, phonophobia, osmophobia or nausea. Aggravating factors included alcohol (within two hours), tiredness, stress and hunger. During a severe attack, the patient would experience polyuria, polydipsia and low mood. On several occasions the pain woke her up at night. Sumatriptan was ineffective for the attacks.

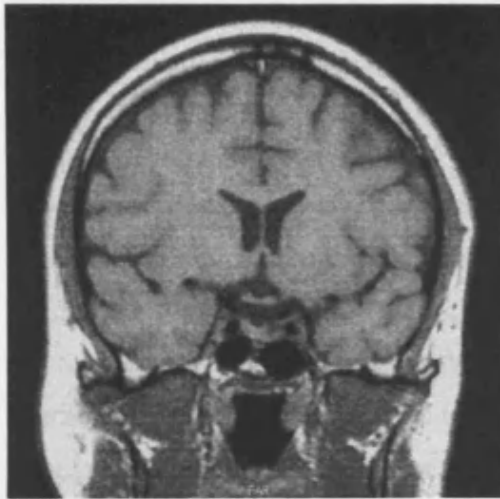
Her serum prolactin was 700 mU/l (27-525 mU/l) and an MRI scan showed a 10mm microadenoma (Figure 30). A diagnosis of a microprolactinoma was made, and she was prescribed bromocriptine 2.5mg. Within 30 minutes of taking the first dose of bromocriptine, she experienced the worst headache she had ever had, leading her to a remark that she felt suicidal due to the severity. The pain was an excruciating left-sided sharp pain with prominent conjunctival injection, lacrimation and ipsilateral ptosis. Simple analgesics, including acetaminophen- (paracetamol) and codeine-containing compounds, did not alleviate the symptoms and the pain lasted for 12 hours. She described the attack as identical to her typical headaches, but without remission. Because of her reluctance to try further dopamine agonists, and persistent

amenorrhoea, headaches and hyperprolactinaemia, transphenoidal removal of the prolactinoma was performed. Post-operatively, she was free from headache. Her serum prolactin normalised and she became pregnant within her menstrual first cycle. She did not experience headache during her pregnancy and her periods returned to normal post-partum.

In 1994 her headaches returned. These were a constant left-sided 'knife-like' pain as before with exacerbations. The exacerbations were the same as before but were more frequent, occurring up to 30 times per day. Several months later, her periods became irregular and she developed galactorrhoea. Her serum prolactin was 1436 mU/l and a repeat MRI showed recurrence of the prolactinoma.

To avoid further surgery, she was given an in-patient trial of cabergoline 250 µg. She again developed an excruciating left-sided headache with cranial autonomic features, within an hour of taking the cabergoline, and lasting 12 hours until being aborted with indomethacin 200mg. Two weeks later she was prescribed quinagolide 250µg. Within 30 minutes she experienced an identical left-sided headache that was of the same intensity and duration as that induced by cabergoline. Previous unsuccessful headache preventative medications include atenolol, pizotifen, and dothiepin. On indomethacin 75mg three times daily for 1 month she had very modest benefit at best. She is currently taking 4800mg gabapentin, which has led to an only very moderate improvement in her symptoms and is being considered for re-exploration of the pituitary fossa.

**Figure 30. Left micro-prolactinoma**



While 1 year of taking bromocriptine, she developed a severe right-sided headache similar to her previous headaches but worse. She continued to take the bromocriptine for 7 1/2 months, and after each dose, she experienced headache. On several occasions, she developed bilateral acromioclavicular joint pain 30 minutes prior to the onset of pain. Since starting the bromocriptine, she developed a second headache. This was a sharp, stabbing occipital pain that took her breath away. It would last no more than a few seconds and occurred several times a week. Her mood became low and the bromocriptine was discontinued.

She was prescribed gabapentin 250mg, because of the problems with bromocriptine. Initially, her headache severity was reduced. On day 14 after the first dose, she developed a sudden onset of right-sided severe headache associated with left hemiplegia. The hemiplegia resolved after 7 days and the gabapentin was stopped, although the headache has persisted at a variable level. Descriptions have been associated with left hemiplegia that resolves leaving each time a background headache. Preventative treatments, including ketoprofen and flunarizine have been unhelpful. A trial of indomethacin produced a dramatic improvement in symptoms.

## 8.2 Case 2

### *Micro-prolactinoma and indomethacin-sensitive headache*

Levy MJ, Matharu MS, Goadsby PJ

Paper ref: European Journal of Neurology 2003;10:1-5

A 40-year-old woman presented in 1997 with severe right-sided headaches. She had experienced headaches during her menarche and has a sister with migraine. She had not experienced headache for over 20 years. The presenting headache was associated with nausea, vomiting, photophobia and phonophobia. Movement would exacerbate the headache. A typical attack would last 3-4 days, and occurred every 2 weeks. During an attack, she experienced ipsilateral conjunctival injection and lacrimation. Triggers included bright lights and sleep disturbance. Two months after the onset of symptoms, she developed secondary amenorrhoea and the frequency of headache increased. Serum prolactin was 2300 mU/L and an MRI scan showed a 9mm microadenoma (Figure 31). She was prescribed bromocriptine 2.5mg.

Within 1 hour of taking bromocriptine, she developed a severe right-sided headache similar to her previous headaches but worse. She continued to take the bromocriptine for 7 ½ months, and after each dose, she experienced headache. On several occasions, she developed bilateral scotomata 30 minutes prior to the onset of pain. Since starting the bromocriptine, she developed a second headache. This was a sharp, stabbing occipital pain that took her breath away. It would last no more than a few seconds and occurred several times a week. Her mood became low and the bromocriptine was discontinued.

She was prescribed cabergoline 250µg, because of the problems with bromocriptine. Initially, her headache severity was reduced. On day 14 after the first dose, she developed a sudden onset of right-sided severe headache associated with left hemiplegia. The hemiplegia resolved after 7 days and the cabergoline was stopped, although the headache has persisted at a variable level. Exacerbations have been associated with left hemiplegia that resolves leaving each time a background headache. Preventative treatments, including tricyclics and flunarizine have been unhelpful. A trial of indomethacin produced a dramatic improvement in symptoms

with abolition of headache within one hour and persistence of the response on 50mg three times daily.

*Acromegaly-associated headache responsive to octreotide but not bromocriptine*

**Figure 31. Right micro-prolactinoma**



She had a one year history of headaches and when characterized by a severe continuous pain in the anterior cilia, there were exacerbating symptoms, nausea, photophobia, vomiting. The headaches were unresponsive to treatment with analgesics. Her history of acromegaly, including nasal bands and enlarged hands, feet and feet. Her serum GH-1 was elevated to 150mIU/L (NR 13-64) and a subsequent oral glucose tolerance test (OGTT) confirmed active acromegaly (mean growth hormone level > 20mIU/L). Her prolactin was 1100mIU/L (NR < 360), total T4 197pmol/L (NR 71-148), TSH 4.10mIU/L (NR 0.3-4.7), LH 11.8U/L (NR 2-10 mid-cycle), FSH 9.3U/L (NR 4-12 mid-cycle). A CT scan showed a pituitary lesion with invasion of the left cavernous sinus.

Despite trans-sphenoidal resection and subsequent radiotherapy, her GH remained elevated and her headache persisted (mean growth hormone 29mIU/L during OGTT). She was given octreotide at a dose of 50mg tds. For the first time since presentation, she noticed an instant improvement in her headaches. The analgesia free, octreotide was completely reproducible, and she was pain-free on a maintenance dose of 150mg tds. She would experience only mild headache one hour before each consecutive dose. An OGTT on octreotide showed a mean growth hormone level of 3.7mIU/L, indicating a good response to medical therapy.

In 1998, she was converted to 30mg bromocriptine LA fortnightly. Although her GH secretion was better controlled (mean value 2.3mIU/L), her headache returned.

### 8.3 Case 3

#### *Acromegaly-associated headache responsive to octreotide but not lanreotide*

Levy MJ, Goadsby PJ, Meeran K

Paper ref: Headache 2003; 43(7): 794-798

A 29 year old lady presented in 1989 with a one year history of headaches and secondary amenorrhoea. Her headaches were characterised by a severe continuous sharp, left retro-orbital pain. In addition to the constant pain, there were excruciating exacerbations associated with a throbbing sensation, nausea, photophobia, phonophobia and aggravation with movement. The headaches were unresponsive to all conventional analgesics.

On examination she had typical features of acromegaly, including broad hands and feet and mild coarsening of her facial features. Her serum IGF-1 was elevated to 150nmol/l (NR 13-64) and a subsequent oral glucose tolerance test (OGTT) confirmed active acromegaly (serum growth hormone nadir > 50mU/l). Her prolactin was 180mU/l (NR < 360), total T4 107nmol/l (NR 71-148), TSH 1.10mU/l (NR 0.3-4.7), LH 11.8U/l (NR 2-10 mid-cycle), FSH 9.3U/l (NR 9-12 mid-cycle). A CT scan showed a pituitary lesion with invasion of the left cavernous sinus.

Despite trans-sphenoidal resection and subsequent radiotherapy, her GH remained elevated and her headache persisted (growth nadir 29mU/l during OGTT). She was given octreotide at a dose of 50µg tds. For the first time since presentation, she noticed an instant improvement in her headache. The analgesia from octreotide was completely reproducible, and she was pain-free on a maintenance dose of 100µg tds. She would experience only mild headache one hour before each octreotide dose. An OGTT on octreotide showed a mean growth hormone level of 3.7mU/l, indicating a good response to medical therapy.

In 1998, she was converted to 30mg lanreotide LA fortnightly. Although her GH secretion was better controlled (mean value 2.5mU/l), her headaches returned.



Subsequent conversion to monthly octreotide LAR led to an immediate improvement in headache symptoms. One week before each injection was due, her headache symptoms would return, and she required additional doses of short-acting octreotide (100mcg) until the next dose of octreotide LAR. Conversion to three-weekly injections of octreotide LAR has led to both excellent growth hormone and headache control. There has been no evidence of tachyphylaxis to the octreotide in terms of headache response for 4 years and her mean GH is now 1.0 mU/l.

#### 8.4 Case 4

##### *Macro-prolactinoma and SUNCT-like headache*

Matharu MS, Levy MJ, Merry RT, Goadsby PJ

Paper ref: Journal of Neurology, Neurosurgery and Psychiatry 2003; 74 (11): 1590-2

A 37-year-old, right-handed, married, man presented with an 11-year history of headaches in December 1998. There were no precipitants at onset. Initially he had 1-2 headaches per month but there was a gradual increase in the frequency over time. In the early to mid 1990s he had bouts of daily attacks alternating with remission periods; he had 2-3 remissions annually with each remission lasting 2-4 weeks. In the late 1990s he had no remissions at all. A typical attack was strictly unilateral on the right. It started as a dull ache in the neck or the occiput and then radiated to the parietal region, vertex, forehead and the temple before becoming centred on the retro-orbital region. The pain was very severe or excruciating and had a squeezing or pressing quality. The usual duration of the pain was 20-30 seconds though the range was 10-60 seconds. The pain came on rapidly, was maintained at a plateau phase and then resolved rapidly. He denied any super-imposed spikes or variations in the severity of the pain. He had 1-6 attacks daily at presentation. The attacks were associated with prominent ipsilateral conjunctival injection and lacrimation, and bilateral facial flushing. He denied all other cranial autonomic features, nausea, vomiting, photophobia, phonophobia or osmophobia. He felt restless during the pain and would often pace up and down. He denied any aura symptoms. Occasionally he had a dull interictal pain that lasted 10-60 minutes. The headaches could be triggered consistently by exercise, particularly cycling, and inconsistently by stressful situations. He denied having any other triggers for the headaches. He was unsure whether there was a refractory period after an attack; the shortest interval between 2 spontaneous attacks was about 3 minutes. He could abbreviate the attack by coughing. There was no past history of headaches.

The patient had previously tried ibuprofen, aspirin, distalgesic and indomethacin 50mgs three times daily for the headaches without any benefit. At presentation, he was not taking any drugs. In the past medical history he had a tonsillectomy in

childhood. His sister was diagnosed as having cancer of the thyroid gland at the age of 21 years. There is no family history of headaches. He is a non-smoker and drinks 25-30 units of alcohol per week.

General and a detailed neurological examination was entirely normal. A magnetic resonance imaging (MRI) scan of the brain revealed a pituitary adenoma on the right extending towards the right cavernous sinus, causing displacement of the right internal carotid artery; the pituitary adenoma also extended into the chiasmatic cistern but did not impinge on the optic chiasm (Figure 32). Formal visual field testing was normal. Blood tests showed that the serum prolactin level was markedly elevated at 10,000 mU/L (normal range, 0 to 635) and testosterone was at the lower end of the normal range (10.1 nmol/L; normal range 9 to 33) with no elevation of luteinizing hormone (LH) or follicle stimulating hormone (FSH), while free thyroxine, thyroid stimulating hormone (TSH), basal cortisol, adrenocorticotrophic hormone (ACTH), oestrogen and growth hormone levels were normal. A diagnosis of macroprolactinoma was made.

At that point the patient was re-interviewed to elicit features of pituitary disease. He reported that, in retrospect, there may have been a slight reduction in libido over the previous few years, although sexual function had otherwise been normal. He had always had scanty beard growth, but otherwise secondary sexual development and characteristics were normal. There was no galactorrhoea and vision was normal.

The patient was started on bromocriptine 2.5mg bd with which the prolactin level normalised within 3 months. The bromocriptine dose was thereafter reduced to 1mg bd. The headaches started improving shortly after starting bromocriptine and resolved completely within 3 months. A repeat MRI scan of the brain in February 2000 showed that the pituitary adenoma had virtually disappeared. The patient continued taking bromocriptine for 18 months but began to develop nausea and nasal blockage; bromocriptine was therefore substituted with cabergoline 1.5mg per week. He has been on cabergoline over the last 18 months on which the prolactin level remains normal and there are no side-effects. He denies having had any further headaches and his libido and energy levels have significantly improved.

**Figure 32. Prolactinoma invading right cavernous sinus**



and a GHRT confirmed this (growth hormone levels 21.6, 24.5, 27.0, 25.9 and 24.7 mIU/L at 0, 30, 60, 90 and 120 minutes respectively). An MRI scan confirmed a pituitary macroadenoma, and the patient underwent a transphenoidal hypophysectomy. Post-operatively, the patient still experienced her headache, and a repeat GHRT showed no reduction in GH but lack of cure (growth hormone levels 10.2, 7.7, 8.1, 9.6-9.3 mIU/L at 0, 30, 60, 90 and 120 minutes respectively). The patient went on to have external radiotherapy, after which growth hormone levels remained  $> 5$  mIU/L and the headache persisted. Because of the disabling nature of the lady's leg, intracranial surgery was performed to remove any residual tumour. Post-operatively, there remained no improvement in headache.

An octreotide infusion (100µg/hour) was given to assess medical response of growth hormone. During this infusion, the patient noticed a dramatic improvement in her headache for the first time for many years. Although the octreotide improved her headache symptoms, there was no reduction in growth hormone during the infusion. Because of the aggressive analgesic response to octreotide, the patient was keen to be started on this agent. With each octreotide injection, the patient experienced an immediate analgesic effect with reduced headache one hour before the next injection. After several months, the patient found that she needed to increase the frequency of octreotide injections as the time interval between headache had become increasingly short. The maximum frequency of octreotide injections was twelve administrations per day. Conversion to monthly injections of octreotide LAk led to recurrence of her persistent headache and she experienced no analgesic effect on this agent.

## 8.5 Case 5

### *Acromegaly and octreotide rebound headache*

Levy MJ and Goadsby PJ

Abstract ref: Endocrine abstracts 2004; 7: P110

A 35 year-old lady presented with secondary amenorrhoea, coarsening of features, growth of hands and feet and persistent unilateral headache. The headache was a featureless continuous left-sided frontal pain. There were occasional exacerbations in the headache, but generally the pain was graded as 6 out of 10 on a visual analogue scale. A clinical diagnosis of acromegaly was suspected, and an oral glucose tolerance test (OGTT) confirmed this (growth hormone levels 21.6, 21.5, 27.0, 25.0 and 24.7 mU/l at 0, 30, 60 90 and 120 minutes respectively. An MRI scan confirmed a pituitary macro-adenoma, and the patient underwent a transphenoidal hypophysectomy. Post-operatively, the patient still complained of headache, and a repeat OGTT showed reduction in GH but lack of cure (growth hormone levels 10.2, 7.2, 8.1, 9.6, 9.3 mU/l at 0, 30, 60 90 and 120 minutes respectively). The patient went on to have external radiotherapy, after which growth hormone levels remained > 5mU/l and the headache persisted. Because of the disabling nature of the headaches, transcranial surgery was performed to remove any residual tumour. Post-operatively, there remained no improvement in headache.

An octreotide infusion (100µg/hour) was given to assess medical response of growth hormone. During this infusion, the patient noticed a dramatic improvement in her headache for the first time for many years. Although the octreotide improved her headache symptoms, there was no reduction in growth hormone during the infusion. Because of the impressive analgesic response to octreotide, the patient was keen to be started on this agent. With each octreotide injection, the patient experienced an immediate analgesic effect with rebound headache one hour before the next injection. After several months, the patient found that she needed to increase the frequency of octreotide injections as the time interval between headache had become increasingly short. The maximum frequency of octreotide injections was twelve administrations per day. Conversion to monthly injections of octreotide LAR led to recurrence of her persistent headache and she experienced no analgesic effect on this agent.

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An MRI of the pituitary fossa revealed residual pituitary tissue ipsilateral to the symptoms of headache (Figure 33). Two courses of gamma-knife radiotherapy were targeted at this residual tumour. Subsequent OGTT revealed cure of her acromegaly (mean growth hormone < 0.5 mU/l) but her headache persisted. She was admitted to the ward for investigation of her headache and withdrawal from octreotide. Numerous abortive agents have been administered in attempt to improve her headache including sumatriptan 6mg s/c, high flow oxygen, intranasal and intravenous lignocaine, intravenous dihydroergotamine and ketamine, and greater occipital nerve injection. Since this time, numerous preventative agents have also been administered, including amitriptyline 75mg, carbamazepine 400mg, gabapentin 5400mg and topiramate 400mg, all without effect.

The patient was readmitted for investigation and a placebo controlled trial of octreotide confirmed that this agent continues to give immediate analgesia. To this date, octreotide is the only agent that has provided any pain relief in this patient. She is currently on the waiting list for an occipital nerve stimulator insertion.

**Figure 33. Residual GH-secreting tumour in right cavernous sinus**



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