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Posterior hypothalamic modulation of light-evoked trigeminal neural activity and lacrimation

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Abstract

Enhanced light sensitivity is a common feature of many neuro-ophthalmic conditions and some chronic headaches. Previously we reported that the bright light-evoked increases in trigeminal brainstem neural activity and lacrimation depended on a neurovascular link within the eye (Okamoto et al., 2012). However, the supraspinal pathways necessary for these light-evoked responses are not well defined. To assess the contribution of the posterior hypothalamic area (PH), a brain region closely associated with control of autonomic outflow, we injected bicuculline methiodide (BMI), a GABA_A receptor antagonist, into the PH and determined its effect on the encoding properties of ocular neurons at the ventrolateral trigeminal interpolaris/caudalis transition (Vi/Vc) and caudalis/upper cervical cord junction (Vc/C1) regions and on reflex lacrimation in male rats under isoflurane anesthesia. BMI markedly reduced light-evoked (> 80%) responses of Vi/Vc and Vc/C1 neurons at 10 min with partial recovery by 50 min after injection. BMI also reduced (> 35%) the convergent cutaneous receptive field area of Vi/Vc and Vc/C1 ocular neurons indicating that both intraocular and periorbital cutaneous inputs were affected by changes in PH outflow. Light-evoked lacrimation was reduced by > 35% at 10 min after BMI, while resting mean arterial pressure increased promptly and remained elevated (> 20 mmHg) throughout the 50 min post-injection period. These results suggested that PH stimulation, acting in part through increased sympathetic activity, significantly inhibited light- and facial skin-evoked activity of ocular neurons at the Vi/Vc and Vc/C1 region. These data provide further support for the hypothesis that autonomic outflow plays a critical role in mediating light-evoked trigeminal brainstem neural activity and reflex lacrimation.

Keywords

trigeminal subnucleus caudalis; light sensitivity; lacrimation; posterior hypothalamus; descending control

Enhanced sensitivity to visible light, often referred to as photophobia or photo-oculodysnia (Lebensohn, 1951; Digre and Brennan, 2012), causes a wide range of ocular sensations that can vary in intensity from mild discomfort to intolerable pain. Among the conditions most

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often associated with photophobia are included: eye injury or inflammation (Custer and Reistad, 2000; Cordero-Coma et al., 2007), blepharospasm (Hallet et al., 2008), retinal dystrophies (Prokofyeva et al., 2011) and some forms of chronic headache (Goadsby et al., 2008; Nosedá and Burstein, 2011).

The diversity of conditions associated with photophobia suggests multiple underlying mechanisms; however, a common feature that may link exposure to light with somatic sensation is the activation of trigeminal sensory nerves (Lebensohn, 1951; Digre and Brennan, 2012). Neuroimaging data revealed specific activation patterns in the trigeminal ganglion (TG) and brainstem in a patient with corneal abrasion by visible light stimulation (Moulton et al., 2009). Exposure to bright light in the rat was sufficient to produce a discrete pattern of Fos-positive neurons at the trigeminal interpolaris/caudalis transition (Vi/Vc) and trigeminal subnucleus caudalis and upper cervical spinal cord junction (Vc/C1) regions (Okamoto et al., 2009), the main terminal zones for trigeminal afferent fibers that innervate the eye (Marfurt and del Toro, 1987; Panneton et al., 2010). Recently we provided evidence that light-evoked trigeminal brainstem neural activity depended on neurovascular events within the eye. Exposure to increasing intensities of light was accompanied by progressive increases in choroidal blood flow that preceded the elevation in trigeminal brainstem neural activity, responses that were prevented by intravitreal injections of vasoactive agents (Okamoto et al., 2012). Early clinical studies found that ocular instillation of epinephrine greatly reduced the sensitivity to bright light in patients with persistent ocular inflammation (Lebensohn, 1934), while light-evoked eye spasm (McCann et al., 1999) and photo-oculodynia and reduced tear secretion in idiopathic ocular pain patients were reversed by cervical sympathetic ganglion blockade (Fine and Digre, 1995). Although these studies suggest sympathetic nervous system involvement in abnormal light sensitivity, little is known about the central pathways that influence sympathetic outflow relevant for light responsiveness in the trigeminal sensory system.

The posterior hypothalamus (PH) has long been associated with sympathetic outflow and pain modulation in humans (Sano et al., 1970, 1975). Short-term PH stimulation in multiple sclerosis patients reduced neuralgic-like pain in the territory of the ophthalmic branch of the trigeminal nerve (Cordella et al., 2009), while long-term stimulation of PH has been used successfully to treat drug-resistant attacks of cluster headache (Leone et al., 2008). Cluster headache (CH) is the most common of the trigeminal autonomic cephalalgias and is characterized by strictly unilateral periorbital pain, ipsilateral lacrimation and conjunctival injection (May et al., 1999, 2006; Leone and Bussone, 2009). Nitrate-induced pain in chronic CH patients evoked a unilateral increase in regional cerebral blood flow in the PH (May et al., 1998), while acute PH stimulation in CH patients caused ipsilateral activation in TG and trigeminal spinal nucleus (May et al., 2006). To address the issue of PH stimulation and light-evoked trigeminal brainstem neural activity and reflex lacrimation, we injected the GABA_A receptor antagonist, bicuculline methiodide (BMI), into the PH of anesthetized male rats. It is well established that PH outflow is under strong GABAergic inhibitory influence and that GABA disinhibition evokes prompt increases in sympathetic activity (Wible et al., 1988) and defense reaction behavior (Shekhar and DiMicco, 1987).

Experimental procedures

The animal protocols were approved by the Institutional Animal Care and Use Committee of the University of Minnesota (USA) and conformed to the guidelines set by The National Institutes of Health guide for the care the use of laboratory animals (PHS Law 99–158, revised 2002). All efforts were made to minimize the number of animals used for experiments and their suffering.

Electrophysiology procedures

Animals

Male rats (270–350 g, Sprague-Dawley, Harlan, Indianapolis, IN) were anesthetized initially with pentobarbital sodium (70 mg/kg, i.p.). Catheters were positioned in the right femoral artery for monitoring blood pressure and jugular vein for drug infusion (gallamine triethiodide, 20 mg/kg/h, at the time of recording). After tracheotomy animals were respired artificially with oxygen-enriched room air and anesthesia was maintained with isoflurane (1.2–2.0%). Expiratory end-tidal CO₂ (3.5–4.5%), mean arterial pressure (MAP, 90–120 mmHg) and body temperature (37°C) were monitored continuously and kept within normal range. Animals were placed in a stereotaxic frame and a portion of the C1 vertebra was removed to expose the lower brainstem and upper cervical dorsal horn. The exposed brainstem surface was bathed in warm mineral oil. Single neurons were recorded from the ventrolateral Vi/Vc transition or superficial laminae (laminae I–II) at Vc/C1 region. Neurons recorded at the Vi/Vc region were approached at an angle of 28° off vertical and 45° off midline, and 1.5–2.0 mm below the brainstem surface. For neurons in superficial laminae at the Vc/C1 region, the electrode was directed at an angle of 43° off vertical, 60° off midline, and the depth of recording was within 250 μm of the dorsal brainstem surface. Extracellular unit activity was recorded using tungsten microelectrodes (5–9 Mohm; Frederick Haer Inc., Bowdoinham, ME) and amplified, discriminated, stored and analyzed offline using a PowerLab interface and LabChart software (AD Instruments, Colorado Spring, CO).

Characterization of light-responsive ocular neurons

The search stimulus consisted of gently swiping a fine camelhair brush across the ocular surface (e.g., cornea surface and conjunctiva). All units included in this study were activated by mechanical stimulation of both the cornea and conjunctiva. Units with a convergent cutaneous receptive field (RF) were classified as wide dynamic range (WDR) or nociceptive specific (NS) based on the responses to a low force von Frey filament (1.2 g) and pinch with blunt forceps as described previously (Hirata et al., 1999, 2004). Neurons with no apparent cutaneous RF were classified as cornea only (CO). The ocular surface was kept moist with artificial tears throughout neural recording experiments.

Light stimulation and experimental design

One neuron was recorded from each animal preparation and only those neurons responsive to light were included for further analysis. Light stimulation was delivered from a thermal-neutral fiber optic halogen source (150W; Cole Parmer, Vernon Hills, IL) positioned 5 cm from the left ocular surface under dim ambient light conditions. Light intensity was measured at the ocular surface with a lux meter (Control Co., Friendswood, TX). High intensity light stimuli (2×10^4 lux, 30 s duration) were presented up to four times in each experiment in a repeated design.

Microinjection of GABA_A receptor antagonist into the posterior hypothalamus

The GABA_A receptor antagonist, bicuculline methiodide (BMI, 1 mM, pH 7.2; Sigma) dissolved in artificial cerebral spinal fluid (aCSF), or vehicle was injected ipsilateral to the stimulated eye via a glass micropipette positioned 4.0–4.6 mm posterior to bregma, 0.5 mm off midline and 7.3–7.6 mm ventral to the cortical surface 10 min prior to light stimulation. Off-target controls received injections of BMI placed 0.5–0.7 mm dorsal-lateral to PH and ipsilateral to light stimulation. The dose of BMI used was shown previously to inhibit dura-evoked second-order trigeminal brainstem neurons and to elevate arterial blood pressure (Bartsch et al., 2004). Injections of BMI or vehicle were delivered slowly over 60 s in a total a volume of 100 nl.

Tear volume

A separate group of male rats (320–400 g, Sprague-Dawley Harlan, Indianapolis, IN) was anesthetized initially with pentobarbital sodium (50 mg/kg, i.p.) and prepared as above for neural recording, but without exposure of the brainstem surface. Tear volume was measured under isoflurane anesthesia (1.0–1.5%) and low ambient light conditions (< 100 lux) and estimated by the increase in wet length of phenol red thread (ZONE-QUICK, Menicon Inc., San Mateo, CA; Hamano et al., 1983) observed to change color (white to red). The thread was placed in contact with the cornea/conjunctiva at the inferior-lateral edge of the cornea/conjunctiva interface immediately after removing excess tears with a fine filter paper. Tear volume then was sampled over 2 min beginning immediately before, during and after 30 s of light stimulation (Okamoto et al., 2010). To assess PH involvement in light-evoked tear formation, high intensity (2×10^4 lux) light stimuli were presented before and 10, 30 and 50 min after BMI (1 mM, 100nl, pH = 7), vehicle or off-target BMI injections via a glass micropipette as described above for neural recording.

Data analysis

Neural recording data were acquired and displayed as peristimulus time histograms of spikes per 1 s bins, exported to a spreadsheet and analyzed off-line. Neural responses were analyzed as a response magnitude (Rmag) for each stimulus period defined as the cumulative sum of spikes for contiguous bins in which the spike count exceeded the mean plus 2 times the standard deviation (SD) of the background activity (Hirata et al., 1999). The total Rmag was calculated for each stimulus period and can be thought of as the “area under the curve”. Neurons were defined as light-responsive if the total Rmag exceeded a value of 10. Response duration was defined as the time interval after stimulus onset until three consecutive bins with a positive spike count occurred above background (initial latency) and until the value of three consecutive bins no longer exceeded the mean + 2SD above background activity. Response latency was defined as earliest time after stimulus onset for which three consecutive 1-s bins exceeded the mean + 2SD of background activity. The high-threshold convergent cutaneous RF area of light-responsive units was determined with a small blunt forceps and mapped onto standardized drawing of the rat face, digitized and quantified by a planimetric method using NIH software (Image J). Average resting MAP was sampled for 60 s prior to and at 10 and 30 min after each light stimulus. Neural responses, cutaneous RF area, tear formation, and MAP were assessed by analysis of variance (ANOVA) corrected for repeated measures. Significant treatment effects were further assessed by Newman-Keuls after ANOVA. The data were presented as mean $\pm$ standard error of mean (SEM) and significance level set at $P = 0.05$.

Histology

At the end of the experiment rats were given a bolus of pentobarbital sodium (100 mg/kg, i.p.) and perfused through the heart with saline followed by 10% formalin and the brain processed to recover the recording sites at the Vi/Vc and Vc/C1 regions or microinjection sites in PH.

Results

General properties of Vi/Vc and Vc/C1 units

A total of 28 light-responsive neurons were recorded at the ventrolateral Vi/Vc transition (NS = 6, WDR = 6, CO = 2) and in superficial laminae at the Vc/C1 (NS = 8, WDR = 6) regions. The recovered recording sites were similar to those reported previously (Hirata et al., 1999, 2004). The average spontaneous activity for Vi/Vc (1.3 ± 0.4 spikes/s, $n = 14$) and

Vc/C1 (1.3 ± 0.5 spikes/s, $n = 14$) units was not different prior to light stimulation ($F_{1, 26} = 0.03$, $P > 0.1$).

Vi/Vc units and PH injections

High intensity light stimulation evoked an increase in Vi/Vc neural activity (Fig. 1A) similar in magnitude to that reported previously (Okamoto et al., 2012). At 10 min after BMI injection into PH ($n = 6$) there was a marked reduction in the magnitude of light-evoked Vi/Vc activity compared with pre-BMI injection values and to other groups ($-92 \pm 3\%$, $F_{3, 32} = 19.7$, $P < 0.001$). This was a consistent finding as all 6 units were inhibited $>80\%$ at 10 min after BMI injection. Light-evoked Vi/Vc unit activity remained depressed at 30 min and recovered slowly by 50 min after BMI. By contrast, injection of vehicle into PH ($n = 4$) or off-target injection of BMI placed dorsolateral to PH ($n = 4$) had no effect compared with pre-injection values (Fig. 2A). Response duration to light stimulation was reduced at 10 min after BMI injection into PH compared with pre-BMI injection values ($-44 \pm 21\%$, $F_{3, 32} = 2.88$, $P < 0.05$), whereas no reduction in duration was seen after vehicle or off-target injections (Fig. 2B). Light-evoked response latency increased transiently at 10 min after BMI injection into PH compared with pre-BMI injection values and to other groups ($283 \pm 190\%$, $F_{3, 32} = 4.7$, $P < 0.05$), whereas vehicle or off-target injections had no effect (Fig. 2C). Background activity of Vi/Vc units was not changed significantly in any of the three treatment groups following injection alone (data not shown). Four of the 6 Vi/Vc units tested with BMI injection into PH also had a convergent receptive field (RF, mean area = 1.4 ± 0.3 cm²). BMI injection into the PH significantly reduced the RF area compared with pre-BMI injection values ($-40 \pm 3\%$, $F_{1, 8} = 39.1$, $P < 0.001$), whereas vehicle or off-target injections had no effect.

Vc/C1 units and PH injections

High intensity light stimulation evoked an increase in Vc/C1 neural activity (Fig. 1B) similar in magnitude to that reported previously (Okamoto et al., 2010). At 10 min after BMI injection into PH ($n = 7$) the magnitude of light-evoked Vc/C1 activity was greatly inhibited compared with pre-BMI injection values and other groups ($-92 \pm 3\%$, $F_{3, 30} = 24.8$, $P < 0.001$). This was a consistent finding as 6 of 7 units were inhibited $>90\%$ at 10 min after BMI injection. In contrast to the response of Vi/Vc units, light-evoked Vc/C1 unit activity recovered completely by 30 min after BMI. Injection of vehicle into PH ($n = 3$) or off-target injection of BMI placed dorsolateral to PH ($n = 4$) had no effect on the Vc/C1 unit responses to light (Fig. 3A). Response duration to light stimulation was reduced marginally at 10 min after BMI injection into PH compared with pre-BMI injections values and other groups ($-50 \pm 29\%$, $F_{3, 30} = 5.8$, $P < 0.005$); however, no reduction was seen after vehicle or off-target injections (Fig. 3B). Light-evoked response latency increased transiently at 10 min after BMI injection into PH compared with pre-BMI injections values and other groups ($590 \pm 240\%$, $F_{3, 30} = 13.4$, $P < 0.001$), whereas vehicle or off-target injections had no effect (Fig. 3C). Background activity of Vc/C1 units did not change significantly in any of the three treatment groups following injection alone (data not shown). All seven Vc/C1 units tested with BMI injection into PH also had a convergent RF (mean area = 1.5 ± 0.2 cm²). BMI injection into the PH significantly reduced the RF area compared with pre-BMI injection values ($-46 \pm 5\%$, $F_{1, 11} = 44.0$, $P < 0.001$), whereas vehicle or off-target injections of BMI had no effect. The locations for recovered injection sites are shown in Fig. 4. No apparent differences were seen in the distribution of injection sites across treatment groups for Vi/Vc and Vc/C1 unit recording experiments (data not shown).

Tear formation

Spontaneous tear formation was low and similar in both eyes (ipsilateral, 3.6 ± 0.4 mm/2 min; contralateral, 3.4 ± 0.4 mm/2min) prior to light stimulation. High intensity light

stimulation increased tear volume more than 2-fold above spontaneous tearing in the ipsilateral eye across all groups prior to drug treatment (8.8 ± 0.5 mm³/2 min, $F_{1, 12} = 118$, $P < 0.001$, $n = 13$), whereas tear volume in the contralateral eye was not affected. BMI injection into PH ($n = 5$) significantly reduced light-evoked tear volume in the stimulated eye over the 50 min test period compared to pre-BMI values ($F_{3, 30} = 6.4$, $P < 0.005$), whereas vehicle ($n = 4$) or off-target injections of BMI ($n = 4$) had no effect (Fig. 5A). The locations for recovered injection sites are shown in Fig. 5B.

Mean arterial pressure (MAP)

Resting and light-evoked MAP were measured in all experiments (resting MAP = 114 ± 2 mmHg, $n = 28$) immediately prior to light stimulation. BMI injection into PH ($n = 13$) caused a prompt increase in MAP that persisted throughout the 50 min experimental period ($F_{3, 73} = 67.4$, $P < 0.001$), whereas vehicle injection in PH ($n = 7$) or off-target injections of BMI ($n = 8$) had no effect (Fig. 6).

Discussion

Acute GABAergic disinhibition by BMI injection into the PH resulted in three novel findings. First, there was a marked reduction in bright light-evoked activity of cornea-responsive neurons recorded at the Vi/Vc transition and in superficial laminae at the Vc/C1 regions. Second, PH disinhibition significantly reduced light-evoked lacrimation. Third, PH disinhibition significantly reduced the RF area of convergent periorbital skin input to Vi/Vc and Vc/C1 neurons. These results indicated that PH had widespread effects on trigeminal sensory processing related to ocular pain and homeostasis.

The PH has long been implicated in modifying pain sensation and autonomic functions in humans (Sano et al., 1970, 1975) and nociceptive responses in animals (Rhodes and Liebeskind, 1978; Millan et al., 1983; Manning and Franklin, 1998; Jeong et al., 2012). However, no previous studies have assessed PH involvement in ocular pain other than those related to clinical management of trigeminal autonomic cephalalgias. Short-term PH stimulation reduced trigeminal neuralgic-like pain in multiple sclerosis patients with the greatest benefit seen in facial skin regions supplied by the ophthalmic branch of the trigeminal nerve; however, effects on ocular sensation were not reported (Cordella et al., 2009). In animal studies involving the ophthalmic branch of the trigeminal nerve, GABAergic disinhibition of PH reduced dural-evoked activity in neurons recorded from deep laminae of Vc (Bartsch et al., 2004, 2005). Although many dura-responsive neurons also had a corneal RF, PH-induced effects on corneal input were not reported.

The mechanisms that mediate PH-induced effects on ocular pain and homeostasis are not certain; however, several general mechanisms may be considered based on known PH efferent projection pathways. First, as described by Jeong et al. (2012), PH-induced inhibition of ocular or cutaneous nociception may involve catechoaminergic mechanisms through the A7 cell group. Synaptic blockade of A7 or intrathecal administration of alpha-2 adrenergic receptor antagonists prevented PH-induced antinociception as measured by tail-flick and paw withdrawal reflexes. Noradrenergic fibers are densely distributed in superficial laminae of Vc (Senba and Tohyama, 1981), while local application of adrenergic agonists inhibits Vc neurons (Sasa et al., 1979; Grudt et al., 1995; Han et al., 2007). Second, PH-induced inhibitory effects may involve the periaqueductal gray - rostral ventromedial medulla system (PAG-RVM), a major endogenous descending pain control system in the brain (Heinricher et al., 2009). The PH projects heavily to the PAG-RVM system (Vertes and Crane, 1996) and RVM stimulation greatly reduces corneal input to caudal trigeminal brainstem nuclear complex (TBNC) neurons (Lovick and Wolstencroft, 1979; Meng et al., 2000). Third, PH efferents may project directly to the Vi/Vc transition and Vc/C1 regions to

inhibit periocular cutaneous or light-evoked responses. Anatomical studies indicate significant projections from PH to the upper cervical spinal cord (Hosoya, 1980; Holstege, 1987). Further, orexin-positive neurons, a peptide neuromodulator synthesized exclusively in neurons of the PH and adjacent hypothalamic regions, also project densely to dorsal horn and intermediolateral regions of the spinal cord (van den Pol, 1999) suggesting a role in sensory processing and autonomic control. In preliminary studies, direct application of orexin A to the dorsal surface at the Vc/C1 region greatly reduced light-evoked responses and periorbital cutaneous RF area of corneal-responsive neurons (unpublished observations, this laboratory). Fourth, as we proposed previously (Okamoto et al., 2010, 2012), a neurovascular mechanism within the eye may be critical in linking exposure to bright light with evoked trigeminal nerve activity and reflex lacrimation. Acute stimulation of PH may inhibit these responses by interrupting autonomic control circuits. The PH (Vertes and Crane, 1996) and adjacent lateral hypothalamus (Goto and Swanson, 2004) project to several central autonomic control regions including the rostral ventrolateral medulla, nucleus tractus solitarius and the superior salivatory nucleus (SSN). The SSN is the major source of preganglionic parasympathetic neurons output to the eye (Neuhuber and Schrod, 2011). Acute PH stimulation in humans increased blood pressure and pulse rate and was often accompanied by pupillary dilatation (Sano et al., 1970). In anesthetized rats (Bartsch et al., 2004, 2005; current study) PH disinhibition caused a prompt elevation in arterial pressure, while similarly in conscious rats, arterial pressure, heart rate and splanchnic sympathetic nerve activity were rapidly increased (Wible et al., 1988). Previously we proposed that changes in choroidal blood flow, which is strongly controlled by the autonomic nervous system (Kur et al., 2012), provides the link between light stimulation and trigeminal nerve activity since it increased in proportion to light intensity, preceded the increase in trigeminal brainstem neural activity and could be prevented by intravitreal administration of phenylephrine, a selective α -adrenergic receptor agonist (Okamoto et al., 2012). Intravitreal injections of norepinephrine also reduced light-evoked Fos expression at the Vc/C1 region (Okamoto et al., 2009). Interestingly, neither intravitreal nor intra-TG injections of phenylephrine affected the convergent cutaneous RF area of Vc/C1 (Okamoto et al., 2010). Similarly, intravitreal injections of phenylephrine or nitric oxide synthase had no effect on the cutaneous RF area of Vi/Vc transition neurons (Okamoto et al., 2012) suggesting that at least some of the PH-induced effects on ocular-related sensory processing was due to a central neural mechanism rather than catecholamine actions on peripheral nerve endings in facial skin or within the eye. It also was not likely that changes in MAP accounted for the PH-induced effects on TBNC neurons since the time course changes in light-evoked neural activity and reflex lacrimation in relation to changes in MAP did not coincide completely. Light-evoked changes in neural activity at the Vi/Vc and reflex lacrimation matched well over time: however, light-evoked changes in Vc/C1 neural activity recovered rapidly by 30 min after BMI injection, whereas MAP rose rapidly and remained elevated throughout the 50 min post-injection period. These results suggested that the underlying mechanisms for PH-induced influence on light-evoked responses were different for Vi/Vc and Vc/C1 neurons and were consistent with the well-established view that centrally mediated changes in autonomic outflow display marked regional and tissue specificity (Janig and McLachlan, 1992; Morrison, 2001).

As discussed above, the central neural mechanisms that mediate PH-induced changes in autonomic outflow and trigeminal neural activity related to ocular functions likely include multiple descending pathways. However, direct ascending pathways from the TBNC to the PH and adjacent hypothalamic regions (Malick and Burstein, 1998; Cavdar et al., 2001; Gauriau and Bernard, 2004) further suggest a close relationship between trigeminal sensory processing and pain modulatory regions of the hypothalamus. In the present and previous studies (Bartsch et al., 2004, 2005) BMI was injected unilaterally into the PH and ipsilateral to the site of recording. The rationale for this approach was based on anatomical as well as

clinical findings. The descending projections from PH to brainstem displayed a predominantly ipsilateral course, although contralateral projections also were seen (Holstege, 1987; Vertes and Crane, 1996). Cluster headache (CH) attacks consist of strictly unilateral pain localized to the periorbital region and accompanied by ipsilateral signs of parasympathetic hyperactivity (e.g., lacrimation, conjunctival injection, increased ocular blood flow) and sympathetic deficit (e.g., ptosis, miosis) relevant to ocular function (Horven et al., 1989; May et al., 2006). Corneal pain thresholds were reduced ipsilateral to the attack in CH patients (Sandrini et al., 1991). Although cutaneous sensitivity to mechanical and thermal stimulation of the cheeks was not different on the affected versus unaffected side or during versus outside a CH attack, no testing was made within the ophthalmic division of the trigeminal nerve (Ladda et al., 2006). Experimentally-induced pain in CH patients resulted in increased regional blood flow in the PH that was strictly ipsilateral to the hemisphere of the spontaneous attacks (May et al., 1998), while acute stimulation of the PH increased regional blood flow in the ipsilateral spinal trigeminal nucleus and TG (May, 2008). Collectively, these studies strongly suggest that the pathways from the PH to autonomic or trigeminal brainstem centers are mainly ipsilateral. Stimulation of the PH ipsilateral to the side of attack has been used successfully to treat drug-resistant CH patients; however, significant treatment effects typically were not seen until days if not weeks of continuous stimulation were delivered (Leone et al., 2008; May, 2008). Quantitative sensory testing under chronic PH stimulation conditions in CH patients revealed increased cold pain thresholds to supraorbital skin stimulation compared to control subjects; however, no tests of ocular sensitivity were reported (Jurgen et al., 2009). Thus, although some aspects of the present study such as PH-induced reductions photosensitivity by trigeminal brainstem neurons and reflex lacrimation following exposure to bright light were consistent with the general effects of PH stimulation in CH patients, the dramatic differences in time to effectiveness suggest additional underlying mechanisms likely contribute to the effects of PH stimulation seen in CH patients.

Conclusions

This study used GABAergic disinhibition by BMI injection to determine the effect of PH outflow on the encoding properties of light-responsive ocular neurons at the ventrolateral Vi/Vc and Vc/C1 regions and on reflex lacrimation. BMI significantly reduced the light-evoked responses and the convergent cutaneous receptive field area of Vi/Vc and Vc/C1 ocular neurons suggesting that PH outflow can modify intra-ocular as well as periorbital cutaneous inputs. Light-evoked lacrimation was reduced after BMI in the eye ipsilateral to the injection. Although the exact mechanisms underlying these effects are not certain, the marked increase resting mean arterial pressure after BMI suggested increased sympathetic activity had a significant role. These data strengthen support for the hypothesis that central neural control of the autonomic nervous system plays a critical role in mediating light-evoked trigeminal brainstem neural activity and reflex lacrimation.

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Abbreviations

aCSF	artificial cerebral spinal fluid
BMI	bicuculline methiodide
CH	cluster headache

MAP	mean arterial pressure
NS	nociceptive-specific
PH	posterior hypothalamus
RF	receptive field
Rmag	response magnitude
Vc	trigeminal subnucleus caudalis
Vc/C1	trigeminal subnucleus caudalis and upper cervical spinal cord junction
Vi	trigeminal subnucleus interpolaris
Vi/Vc	trigeminal subnucleus interpolaris/caudalis transition
WDR	wide dynamic range

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Highlights

- 1) GABAergic disinhibition of posterior hypothalamus modifies ocular sensory processing.
- 2) Posterior hypothalamus outflow inhibits light-evoked trigeminal caudalis neural activity and lacrimation.
- 3) Posterior hypothalamus outflow inhibits facial skin input to trigeminal caudalis neurons with ocular receptive fields.
- 4) Posterior hypothalamus stimulation elevates mean arterial pressure.

Figure 1.

Peristimulus time histograms displaying examples of the effects of PH injection of BMI on bright light-evoked Vi/Vc (A) and Vc/C1 (B) unit activity. BMI was injected 10 min before light stimulation (20k lux, 30 s). Bar = 30s (light stimulation). Abbreviations: FR, firing rate.

Figure 2.

Local injection of BMI into the PH ipsilateral to light stimulation decreased light-evoked Vi/Vc unit activity. Mean values of response magnitude (Rmag, **A**) and response duration (**B**) and latency (**C**). * $P < 0.05$, ** $P < 0.01$ versus pre-BMI values (0 min). a = $P < 0.05$, b = $P < 0.01$ versus other groups. Solid bar = PH-BMI (n = 6), open bar = PH-vehicle (n = 4), hatched bar = off-target injection of BMI (n = 4).

Figure 3.

Local injection of BMI into the PH ipsilateral to light stimulation decreased light-evoked Vc/C1 unit activity. Mean values of response magnitude (Rmag, **A**) and response duration (**B**) and latency (**C**). ** $P < 0.01$ versus pre-BMI values (0 min), b = $P < 0.01$ versus other groups. Solid bar; PH-BMI (n = 7), open bar = PH-vehicle (n = 3), hatched bar = off-target injection of BMI (n = 4).

Figure 4.

A. Injection sites in the PH: solid triangles = Vi/Vc units, BMI injection (n = 6); open triangles = Vi/Vc units, vehicle injection (n = 4); solid circles = Vc/C1 units, BMI injection (n = 7), open circles = Vc/C1 units, vehicle injection (n = 3). Off-target injections of BMI are represented by solid (Vi/Vc units; n = 4) and open (Vc/C1 units; n = 4) squares. **B.** An example of a PH BMI injection site (arrow). Abbreviations: F, fornix; LHA, lateral hypothalamus; PH, posterior hypothalamus; V3, third ventricle.

Figure 5.

Disinhibition of PH prevents bright light-evoked tear formation. **A.** Ipsilateral injection of BMI into the PH blocks light-evoked tear formation. * $P < 0.05$, ** $P < 0.01$ versus pre-BMI value (0 min). ^a $P < 0.05$ versus other groups. **B.** Injection sites in the PH: solid circles = BMI injection ($n = 5$); open circles = vehicle injection ($n = 4$), triangles = off-target injection of BMI ($n = 4$). Abbreviations: F, fornix; LHA, lateral hypothalamus; PH, posterior hypothalamus; V3, third ventricle.

Figure 6.

Disinhibition of the PH increases resting mean arterial pressure (MAP). Ipsilateral injection of BMI, but not vehicle and off-target injection of BMI, increases resting MAP. * $P < 0.05$, ** $P < 0.01$ versus 0 min; a = $P < 0.05$, b = $P < 0.01$ versus vehicle and off-target injection of BMI group. Solid bar = PH-BMI (n = 13), open bar = PH-vehicle (n = 7), hatched bar = off - target injection of BMI (n = 8).