Published in final edited form as:

Psychol Med. 2011 February ; 41(2): 385–393. doi:10.1017/S0033291710000838.

Controlled cross-over study in normal subjects of naloxone-preceding-lactate infusions; respiratory and subjective responses: relationship to endogenous opioid system, suffocation false alarm theory and childhood parental loss

M. Preter^{1,*}, S. H. Lee², E. Petkova³, M. Vannucci⁴, S. Kim⁵, and D. F. Klein⁶

¹Department of Psychiatry, Columbia University, and New York State Psychiatric Institute, New York, NY, and Department of Neurology, State University of New York, Downstate Medical Center, Brooklyn, NY, USA

²The Nathan S. Kline Institute for Psychiatric Research, Orangeburg, NY, USA

³Department of Child and Adolescent Psychiatry, New York University School of Medicine, New York, USA

⁴Department of Statistics, Rice University, Houston, TX, USA

⁵Department of Biostatistics, University of Michigan, Ann Harbor, MI, USA

⁶Phyllis Green and Randolph Cowen Institute for Pediatric Neuroscience, Department of Child and Adolescent Psychiatry, New York University Langone Medical Center; The Nathan S. Kline Institute for Psychiatric Research; Department of Psychiatry, College of Physicians and Surgeons, Columbia University, New York, NY, USA

Abstract

Background—The expanded suffocation false alarm theory (SFA) hypothesizes that dysfunction in endogenous opioidergic regulation increases sensitivity to CO₂, separation distress and panic attacks. In panic disorder (PD) patients, both spontaneous clinical panics and lactate-induced panics markedly increase tidal volume (TV), whereas normals have a lesser effect, possibly due to their intact endogenous opioid system. We hypothesized that impairing the opioidergic system by naloxone could make normal controls parallel PD patients' response when lactate challenged. Whether actual separations and losses during childhood (childhood parental loss, CPL) affected naloxone-induced respiratory contrasts was explored. Subjective panic-like symptoms were analyzed although pilot work indicated that the subjective aspect of anxious panic was not well modeled by this specific protocol.

Method—Randomized cross-over sequences of intravenous naloxone (2 mg/kg) followed by lactate (10 mg/kg), or saline followed by lactate, were given to 25 volunteers. Respiratory

physiology was objectively recorded by the LifeShirt. Subjective symptomatology was also recorded.

Results—Impairment of the endogenous opioid system by naloxone accentuates TV and symptomatic response to lactate. This interaction is substantially lessened by CPL.

Conclusions—Opioidergic dysregulation may underlie respiratory pathophysiology and suffocation sensitivity in PD. Comparing specific anti-panic medications with ineffective anti-panic agents (e.g. propranolol) can test the specificity of the naloxone + lactate model. A screen for putative anti-panic agents and a new pharmacotherapeutic approach are suggested. Heuristically, the experimental unveiling of the endogenous opioid system impairing effects of CPL and separation in normal adults opens a new experimental, investigatory area.

Keywords

Affective neuroscience; childhood parental loss (CPL); endogenous opioids; panic disorder pathophysiology

Introduction

Sodium lactate infusions and CO₂ inhalation regularly produce panic attacks in patients with panic disorder (PD) (Gorman *et al.* 1984; Liebowitz *et al.* 1984a; Papp *et al.* 1993; Keck & Strohle, 2005). Distinctively, in most panic patients, both spontaneous and lactate-induced panic attacks produce subjective dyspnea (air hunger) and marked objective increases in tidal volume (TV) and respiratory frequency (Goetz *et al.* 1993; Martinez *et al.* 1996). Healthy control subjects usually respond to a sodium lactate infusion with minor, but definite, hyperventilation (Liebowitz *et al.* 1985). This has been replicated in a rat model (Olsson *et al.* 2002).

Sodium lactate infusion causes a metabolic alkalosis. Therefore, a ventilatory decrease, producing increased CO₂ retention, would be expected to maintain normal blood pH homeostatically. However, the opposite occurs, indicating a stimulatory respiratory effect of lactate, despite metabolic alkalosis.

Because an increase in the partial pressure of carbon dioxide (pCO₂), in addition to increasing blood and brain lactate levels, signals impending asphyxiation, Klein (1993) advanced the integrative suffocation false alarm theory (SFA) of PD. The SFA posits that many seemingly spontaneous panic attacks are due to a dysfunctional, hypersensitive monitor for indications of impending suffocation. Enclosed still air, perceiving labored breathing and sighing inspirations, and also rising pCO₂ levels, such as occur in deepening sleep, premenstrual and post-partum periods, may trigger this specific alarm system. The alarm instigates a flight to help (Preter & Klein, 1998, 2008).

This modular view of multiple adaptive brain systems evolved to deal with specific dangers is supported by Corfield *et al.* (1995). In a positron emission tomography (PET) study of CO₂-stimulated breathing in normal subjects, activation of limbic and specific cerebellar structures was demonstrated. Similarly, neuroimaging data from PET and functional magnetic resonance imaging (fMRI) implicate the cerebellum in the hypercapnic production

of air hunger (Brannan *et al.* 2001; Parsons *et al.* 2001; Evans *et al.* 2002), a ‘compelling primal emotion like severe thirst’ (Liotti *et al.* 2001).

Pathological childhood separation anxiety frequently antecedes PD (Klein, 1964; Gittelman & Klein, 1984; Lipsitz *et al.* 1994; Biederman *et al.* 2006). It was noted that opioids decrease respiratory drive by blocking CO₂ sensitivity. Opioids also decrease separation anxiety whereas the opioid antagonist naloxone was reported to exacerbate separation anxiety (Kalin *et al.* 1988). Therefore, fluctuating opioidergic deficiency might underlie both separation anxiety and a hypersensitive suffocation monitor (Preter & Klein, 1998, 2008).

To explore opioidergic relevance, Liebowitz *et al.* (1984b) infused lactate, and subsequently naloxone preceding lactate, in panic patients with previous lactate sensitivity. However, the low naloxone pretreatment dose chosen (maximum 37.5 mg) did not intensify the lactate infusion response. It was then hypothesized that if clinically symptomatic patients were in a state of opioidergic deficiency, naloxone receptor blockade would have little effect. Conversely, in normal subjects, naloxone, by impairing opioidergic protection, should intensify lactate effects. An open pilot study (Sinha *et al.* 2007) found that, in eight of 12 normal subjects, naloxone infusion (ranging from an initial 0.5 mg/kg to a maximum of 2 mg/kg), followed by intravenous (i.v.) lactate, caused significant TV increments, resembling clinical and lactate-induced panics. This seemed to be dose dependent. The four subjects receiving the maximum dose all manifested marked TV increments. Furthermore, four recalled subjects had no respiratory effect from naloxone alone. However, subjective panic reports were rare.

To extend these pilot findings, we conducted the present controlled, randomized experimental study. We also explored whether life events might influence our findings. Many animal data link endogenous opioid systems to separation and loss (Panksepp, 2003; Preter & Klein, 2008), and specifically to human panic (as opposed to fear). We investigated whether childhood parental loss (CPL) might cause a sub-clinical impairment of the opioidergic system. If so, a decrease in the anticipated robust respiratory response to the naloxone+lactate challenge would occur. Whether this opioidergic downregulation might induce a change in pain perception was also explored.

Previous work (Goetz *et al.* 1993; Martinez *et al.* 1996) in panic patients indicated that the major respiratory effect of apparently spontaneous panic was an increase in TV. Respiratory rate (RR) effects were relatively minor but might reinforce a minute volume (MV) increment (because MV is the multiple of TV and RR). Our primary hypothesis was that, in normal subjects given lactate infusions, randomized pretreatment with naloxone would cause larger increases in TV, RR and MV than pretreatment with saline. Our secondary hypotheses were that (1) pretreatment with naloxone, compared to saline, would increase subjective respiratory symptomatology (such as air hunger) during i.v. lactate, and (2) a history of CPL would interact with the respiratory measures, so that the amplifying effect of naloxone on the lactate infusion would be decreased. Finally, we explored pretreatment with naloxone with regard to reports of pain.

Method

Sample

Healthy volunteers were recruited by advertisement. The age range for female subjects was 18–40 years, to exclude menopausal women; the male age range was 18–50 years. Women were only tested immediately post-menses, after a negative pregnancy test. Subjects underwent a physical examination, routine blood and urine tests, and an electrocardiogram (ECG). The study was approved by the Institutional Review Board (IRB) of the New York State Psychiatric Institute and all participants provided written informed consent.

Exclusion criteria were current or past DSM-IV-defined psychiatric illness; clinically significant heart, lung or neurological condition; clinically significant irritable bowel syndrome, fibromyalgia, thyroid disease, premenstrual syndrome (PMS); menopause; application pending for medical disability; smoking; any sleep disturbance; a positive pregnancy test.

A trained telephone interviewer performed the initial screening, using a modified SCID (Spitzer *et al.* 1992). Suitable volunteers were diagnostically interviewed by the research psychiatrist (M.P.). Eligible volunteers were further interviewed about potentially significant individual and family antecedents and frequent co-morbidities of panic, such as near-suffocation, pulmonary disease and migraine. Recent and childhood loss and separation events (parental divorce or death, childhood abuse) were reviewed specifically.

Protocol

We initially used a three-arm parallel groups design with 30 subjects randomized to naloxone + lactate (NL), 30 to naloxone + saline (NS), and 10 to saline + lactate (SL), with an interim analysis after 50% ($n = 35$ subjects) of the target sample of 70 [reduced from 90 due to National Institute of Mental Health (NIMH) budget cuts]. This interim analysis was performed by both Columbia University (E.P.) and independent statisticians (M.V., S.H.L., S.K.). The project researchers remained blind.

The interim results showed that the concern that naloxone by itself might cause a significant ventilatory increment was unfounded (details on request). This was entirely consonant with the pilot trial by Sinha *et al.* (2007). Therefore, the NS condition was terminated. However, the NL *v.* SL contrast was nonsignificant because the SL sequence had a much greater magnitude and between-subject TV variability than previous experience suggested. Therefore, the protocol was changed, and IRB approved, to a randomized cross-over of NL and SL, administered on two different days.

Randomization

Given subject recruitment difficulties, seven subjects already tested under NL or SL agreed to be recalled, gave consent and were tested under the alternative condition. Six subjects previously NS tested and 14 new volunteers were randomized to the NL and SL sequences. However, two of the 14 randomized to (NL, SL) did not return after the first infusion, restricting this data analysis to 25 subjects with complete data. Although this procedure

enabled only single observer blinding for recalled subjects, the data were generated by objective LifeShirt measures (Grossman, 2004), or self-reports.

Procedures

Subjects reported to the Biological Studies Unit at 0800 or 1200 h after overnight abstinence from food, liquids, caffeine and other stimulants. The LifeShirt apparatus was fitted. This non-invasive ambulatory, Food and Drug Administration (FDA)-approved monitor uses inductive plethysmography (IP) sensors to record respiratory and cardiac function (Grossman, 2004). Sensors include an accelerometer to record postural shifts. A continuous, spirometrically calibrated measurement of TV was recorded, in addition to RR and an ECG.

After negative urine toxicology, and a negative pregnancy test for females, an intravenous cannula was inserted into each arm's antecubital vein; one for infusions, the other for drawing blood.

Testing consisted of four phases. During phase 1, baseline measurements were recorded for 30 min. In phase 2 (first infusion), subjects received naloxone (2 mg/kg) or saline for 3 min based on the randomization assignment, followed by a 17-min rest period. In phase 3 (second infusion), subjects were switched to 0.5 M sodium lactate, 10 ml/kg, administered i.v. over 20 min. This replicates past lactate administrations exactly. Phase 4 (120 min) was the recovery/washout stage, with continued physiological and symptomatic safety monitoring of subjects, but too irregular to provide research data.

The research psychiatrist (M.P.) was present during the entire procedure to monitor physiological output parameters (blood pressure, pulse, respiration) and, as indicated, to confirm abnormal measures manually. The procedure was audio-visually recorded for quality control and further exploration.

Measures

Objective respiratory measurements (digitized 50 times per second) were used to compute the median TV, RR (breaths/min) and MV (product of TV and RR). The two treatment conditions (NL v. SL) were compared during the last 17 min of the lactate infusion. Subjective sensitivity was measured by the modified 17-item Acute Panic Inventory (API; Dillon *et al.* 1987; Papp *et al.* 1997), through non-verbal symptom reports. The Borg Breathlessness Scale (Borg, 1982) is the standard dyspnea measure used by pulmonologists. Subjective scales were administered at baseline, and before and after each infusion period. The 'Panic' measure was the sum of eight API items (each scored from 0 to 3: API2/Fear of Dying; API3/Impending Doom; API5/Air Hunger; API6/Difficulty in Breathing; API9/Dizziness; API10/Confusion; API12/Feeling Detached from All or Part of Your Body; and API13/Sweating). 'Pain' (self-report) was a dichotomous measure indicating whole-body or head pain.

Statistical analysis

The enormous amount of functional data collected required preprocessing (trace artifact correction, baseline adjustment, thinning and noise removal by wavelet decomposition)

before statistical comparisons could be made between NL and SL with respect to the respiratory response curves over time. A special analytic method was developed (Lee *et al.* 2008) to test the dominance of one mean curve over another, incorporating the correlation between repeated observations. The results, presented in *Biometrics* (Lee *et al.* 2008), a technical statistical journal, were in keeping with our respiratory hypotheses. However, the requirements of a largely non-statistical readership led us to this simpler analysis and exposition.

The repeated measurements of median TV, RR and MV were plotted for each subject against time, assessing the shape of the trajectories under NL and under SL during the 17 min before the end of the lactate infusion. The repeated measures were modeled as functions of time (with linear and quadratic terms), experimental condition (NL or SL) and its interaction with time, while controlling for the baseline value of each measure. To account for the correlation between repeated observations on the same subjects, mixed-effect models were used with random intercepts, slopes and curvature for each subject-by-condition, in addition to random subject effects. Non-significant interaction terms were removed one by one, preserving the hierarchical principle that, if a higher-order term is significant, the lower-order terms that comprise it remain in the model. NL *v.* SL contrasts are based on these final models. The effect of CPL on NL *v.* SL contrasts was assessed, by adding terms for CPL plus CPL interactions and using the model selection procedure outlined above.

The effect of pretreatment with naloxone on the 'Panic' and 'Pain' measures was similarly assessed by generalized linear models (GLMs). 'Pain', a dichotomous measure, was modeled with logit link; 'Panic' (a sum of eight API measures, scored 0–3) was modeled with a gamma link. The distribution of the sum did not deviate from gamma distribution. To account for the potential correlation between the values of the same subject's outcomes under NL and SL conditions, mixed-effect GLMs were used. The effect of CPL on the difference between NL and SL with respect to subjective measures was assessed, similar to the effects on respiratory measures.

NL and SL challenges were randomly administered on two different days. The effect of order (NL first *v.* SL first) was not significant and is not reported here.

Although our hypotheses were directional, two-tailed tests were performed using $\alpha = 0.05$. All analyses were performed using SAS 9.1.3 (SAS Institute Inc., USA); the mixed-effects models were fit using Proc MIXED, GLIMMIX and GENMOD.

Results

NL/SL effect on respiratory measures

The final models for TV, RR and MV measures are reported in Table 1. Fig. 1 represents model-based mean trajectories for TV, RR and MV over the last 17 min of the lactate infusion. As predicted, TV during lactate infusion is higher during NL than during SL, and this effect starts almost immediately. The quadratic effects indicate that the initial increments plateau for both SL and NL groups. RR during lactate is also higher for NL than SL, but this appears in the interaction of treatment by time; that means that the NL effect on

RR is initially negligible but steadily increases over time. Note that simply analyzing for a mean effect over the entire infusion would have missed this effect. Finally, MV during lactate under both NL and SL increases steadily during the infusion and at all times is uniformly larger for NL than SL.

The initial clinical interview prospectively identified those exposed to CPL. In keeping with prior work (Brown & Harris, 1978; Agid *et al.* 1999), we defined CPL as marked separations and disruptions (parental death or divorce) occurring prior to age 10. Thirteen probands (six females, seven males) had no history of CPL. Twelve (five females, seven males) had such antecedents. Eleven of the 12 CPL subjects (92%) had experienced parental separation or divorce in early childhood, one subject had lost his mother at age 4. The effects of CPL are summarized in Table 2.

As Table 2 shows, there is no simple CPL effect: CPL only becomes significant by its interaction with condition (NL *v.* SL). Fig. 2 shows the final models for TV, RR and MV. Within each CPL group, NL is greater than SL. This difference is larger within the non-CPL group. In fact, there was no difference between NL and SL in the CPL group with respect to RR. As visual inspection suggests, for TV during NL exposure, the group without CPL was well above the other three conditions: a *post-hoc* analysis was performed contrasting the non-CPL group on NL against all other conditions showing very significant effects ($p < 0.0001$).

Secondary hypotheses: NL *v.* SL effect on subjective reports

Across all groups, there were no significant differences in subjective measures (API, Borg, physical pain) under NL *v.* SL. Taking CPL status into account did not affect these analyses.

Discussion

Normal subjects, usually relatively insensitive to the TV effects of lactate infusion, in this study, given opioid antagonist pretreatment, developed TV and RR increments resembling those occurring in both spontaneous clinical panic attacks and in panic patients who panic during lactate infusions (Gorman *et al.* 1984; Liebowitz *et al.* 1984a; Papp *et al.* 1993). The hypothesis that a functioning endogenous opioid system buffers normal subjects from the behavioral and physiological effects of lactate is consonant with these results.

In this controlled, randomized study, we confirm the findings of the open pilot trial by Sinha *et al.* (2007), the only other naloxone-preceding-lactate study in normal subjects. As in Sinha's study, subjective reports of panic-like symptoms were insignificant. The absence of major measurable subjective responses to the naloxone-lactate challenge previously reported by Sinha *et al.* clearly limits inferences concerning affective reactivity. Subject-endorsed APIs were fairly low, under both NL and SL, and there was little correlation between respiratory API indices and TV increment. Perhaps even higher naloxone doses would have been required for a subjective panic attack to occur. Infusion speed might have played a role. We administered lactate as a slow drip over 20 min. In Sinha *et al.* (2007), the one subject receiving a high-speed lactate infusion, because of an inadvertently wide-open spigot, had a full-blown panic attack.

Earlier explorations of the endogenous opioid system using naloxone showed inconsistent results. Weinberger *et al.* (1985) did not find any effect of naloxone pretreatment on ventilatory responsiveness to 7% CO₂ rebreathing, but the naloxone dose was only 10 mg. Subjective responses were not reported. By contrast, Pickar *et al.* (1982), using 3–6 mg/kg naloxone in normal subjects, found ‘(t)he behavioral effects of high-dose naloxone comprised a rather distinct clinical syndrome characterized by irritability, anxiety, sadness, and confusion.’ This was accompanied by significant dose-dependent increments in RR.

To our knowledge, this is the first time that the prolonged physiological effects of actual separations and losses during childhood (i.e. parental death, parental separation or divorce) on the endogenous opioid system of healthy adults have been objectively shown in an experimental setting. The presence or absence of CPL antecedents determined the response to the naloxone–lactate probe. A history of CPL decreased the NL effect. The import of these findings is that analyses attempting to relate CPL to other baseline variables may well fail because CPL impact may be specific to challenges to the endogenous opioid system. Furthermore, based on our analysis of CPL *v.* non-CPL, the NL *v.* SL contrast across groups was largely a function of non-CPL reactivity to lactate if preceded by naloxone. The NL non-CPL group was most sensitive to the respiratory stimulation effect of lactate when preceded by opioid blockade.

It should be emphasized that these CPL effects were apparent in ‘normal’ subjects. In a society where divorce rates approach 50%, the results raise the question whether current psychiatric classification and diagnostic scales are sensitive enough to detect CPL effects. This also applies to developing societies such as China, where the massive migration of mostly young individuals from the countryside to urban areas has left behind approximately 30 million small children (Liu *et al.* 2009).

CPL is a risk factor for adult anxious and depressive psychopathology (Kendler *et al.* 1992; Bandelow *et al.* 2002). However, its detrimental long-term effect is not limited to psychiatric illness (Shonkoff *et al.* 2009). Using criteria similar to ours, the Adverse Childhood Experiences (ACE) Study, a Centers for Disease Control and Prevention (CDC)-supported prospective cohort study of 16908 adults, found a significant relationship between CPL and premature death in adulthood (Brown *et al.* 2009). Retrospective (e.g. Juang *et al.* 2004; Kopec & Sayre, 2005) and prospective longitudinal data (Fearon & Hotopf, 2001; Harter *et al.* 2003; Katerndahl, 2008; Jones *et al.* 2009) link family disruption, physical abuse, separation and maternal loss in early life to chronic physical pain in adulthood. Whether the NL *v.* SL probe has a differential effect on pain perception and physiological pain measures, and whether CPL status modifies this interaction, should be explored. Unfortunately, our exploratory pain measure was limited to a single item and, in retrospect, was clearly inadequate.

CPL as related to childhood separation anxiety, adult PD and suffocation hypersensitivity was studied by Battaglia *et al.* (2009). In a large sample of twins from Norway, CPL accounted in no small part for ‘the covariation between separation anxiety in childhood, hypersensitivity to CO₂ (as indexed by the anxiety response to a 35% CO₂/65% O₂

mixture), and panic disorder in adulthood'. Note that in Battaglia's study, CPL increased reactivity to the 35% CO₂ probe.

The inclusion of a subgroup of single-blind, recalled subjects is methodologically suspect because recalled subjects have different expectancies from newly recruited subjects. Furthermore, the unblinded experimenter may have subtly changed the environment, although this is somewhat mitigated by our objective measurements. However, given the exigencies of funding and timing, such response bias could not be ascertained.

Our normal, NL-treated controls resemble non-fearful panic patients, an apparent contradiction. The DSM-IV (APA, 1994) criteria for a panic attack require the experience of intense anxiety or discomfort. The absence of anxiety or fear does not preclude panic. Kushner & Beitman (1990) extensively documented the non-fearful panic phenomenon. Epidemiological studies (Katon, 1986) demonstrated that patients with PD often seek non-psychiatric treatment because the clinical presentation often comprises only somatic symptoms. The effect of CPL antecedents on this phenomenon requires further attention.

Testing the specificity of the naloxone–lactate model of clinical panic requires double-blind investigation whether specific anti-panic drugs, but not panic-irrelevant drugs, block this effect. If found, this has practical and heuristic implications. First, there is currently no specific, screening method for testing putative anti-panic drugs except by the experimental treatment of PD patients. The naloxone + lactate effect in normal humans may afford such a screening method, and may be extended to preclinical studies. Second, these data offer heuristic support for the theory that an opioidergic dysfunction is the pathophysiological mechanism underlying PD. If so, the appropriateness of opioidergic therapeutic agents comes into question. The use of morphine or other simple agonists would probably be rejected for fear of inducing addiction, although the evidence for addiction during indicated medical treatment is sparse. However, recent work with opioidergic mixed agonist-antagonists (Gerra *et al.* 2006; Wallen *et al.* 2006), such as buprenorphine, may be relevant. The concern about addiction would be mitigated by the fact that higher doses become receptor blockers rather than agonists. Positive results would foster investigations into basic molecular mechanisms. For instance, we note that the dose of naloxone used in our study (2 mg/kg) substantially exceeds that needed for μ opioid receptor (MOR) blockade (Sluka *et al.* 1999), suggesting a role for the δ opioid receptor (DOR). This could spark interest in the development of specific DOR agonists suitable for human use. Currently, such agents have not been developed, although agents suitable for animal use are available.

Acknowledgments

We thank our subjects, and M. De La Nuez and A. Karyat for their assistance with this study. Dr Vannucci is supported by NSF (DMS-0605001) and by NIH (R01 HG003319-01); Dr Klein by NIMH (MH-30906 and MH-067749).

References

- Agid O, Shapira B, Zislin J, Ritsner M, Hanin B, Murad H, Troudart T, Bloch M, Heresco-Levy U, Lerer B. Environment and vulnerability to major psychiatric illness: a case control study of early

- parental loss in major depression, bipolar disorder and schizophrenia. *Molecular Psychiatry*. 1999; 4:163–172. [PubMed: 10208448]
- APA. *Diagnostic and Statistical Manual of Mental Disorders*. 4th. American Psychiatric Association; Washington, DC: 1994.
- Bandelow B, Spath C, Tichauer GA, Broocks A, Hajak G, Ruther E. Early traumatic life events, parental attitudes, family history, and birth risk factors in patients with panic disorder. *Comprehensive Psychiatry*. 2002; 43:269–278. [PubMed: 12107864]
- Battaglia M, Pesenti-Gritti P, Medland SE, Ogliari A, Tambs K, Spatola CA. A genetically informed study of the association between childhood separation anxiety, sensitivity to CO₂, panic disorder, and the effect of childhood parental loss. *Archives of General Psychiatry*. 2009; 66:64–71. [PubMed: 19124689]
- Biederman J, Petty CR, Faraone SV, Hirshfeld-Becker DR, Henin A, Brauer L, Kaufman B, Rosenbaum JF. Antecedents to panic disorder in nonreferred adults. *Journal of Clinical Psychiatry*. 2006; 67:1179–1186. [PubMed: 16965194]
- Borg GA. Psychophysical bases of perceived exertion. *Medicine and Science in Sports and Exercise*. 1982; 14:377–381. [PubMed: 7154893]
- Brannan S, Liotti M, Egan G, Shade R, Madden L, Robillard R, Abplanalp B, Stofer K, Denton D, Fox PT. Neuroimaging of cerebral activations and deactivations associated with hypercapnia and hunger for air. *Proceedings of the National Academy of Sciences USA*. 2001; 98:2029–2034.
- Brown DW, Anda RF, Tiemeier H, Felitti VJ, Edwards VJ, Croft JB, Giles WH. Adverse childhood experiences and the risk of premature mortality. *American Journal of Preventive Medicine*. 2009; 37:389–396. [PubMed: 19840693]
- Brown, GH.; Harris, TO. *The Social Origin of Depression: A Study of Psychiatric Disorder in Women*. Free Press; New York: 1978.
- Corfield DR, Fink GR, Ramsay SC, Murphy K, Harty HR, Watson JD, Adams L, Frackowiak RS, Guz A. Evidence for limbic system activation during CO₂-stimulated breathing in man. *Journal of Physiology*. 1995; 488:77–84. [PubMed: 8568667]
- Dillon DJ, Gorman JM, Liebowitz MR, Fyer AJ, Klein DF. Measurement of lactate-induced panic and anxiety. *Psychiatry Research*. 1987; 20:97–105. [PubMed: 3575564]
- Evans KC, Banzett RB, Adams L, McKay L, Frackowiak RS, Corfield DR. BOLD fMRI identifies limbic, paralimbic, and cerebellar activation during air hunger. *Journal of Neurophysiology*. 2002; 88:1500–1611. [PubMed: 12205170]
- Fearon P, Hotopf M. Relation between headache in childhood and physical and psychiatric symptoms in adulthood: national birth cohort study. *British Medical Journal*. 2001; 322:1145. [PubMed: 11348907]
- Gerra G, Leonardi C, D'Amore A, Strepparola G, Fagetti R, Assi C, Zaimovic A, Lucchini A. Buprenorphine treatment outcome in dually diagnosed heroin dependent patients: a retrospective study. *Progress in Neuropsychopharmacology and Biological Psychiatry*. 2006; 30:265–272.
- Gittelman R, Klein DF. Relationship between separation anxiety and panic and agoraphobic disorders. *Psychopathology*. 1984; 17(Suppl. 1):56–65. [PubMed: 6369368]
- Goetz RR, Klein DF, Gully D, Kahn J, Liebowitz M, Fyer A, Gorman JM. Panic attacks during placebo procedures in the laboratory: physiology and symptomatology. *Archives of General Psychiatry*. 1993; 50:280–285. [PubMed: 8466389]
- Gorman JM, Askanazi J, Liebowitz MR, Fyer AJ, Stein J, Kinney JM, Klein DF. Response to hyperventilation in a group of patients with panic disorder. *American Journal of Psychiatry*. 1984; 141:857–861. [PubMed: 6428243]
- Grossman P. The LifeShirt: a multi-function ambulatory system monitoring health, disease, and medical intervention in the real world. *Studies in Health Technology and Informatics*. 2004; 108:133–141. [PubMed: 15718639]
- Harter MC, Conway KP, Merikangas KR. Associations between anxiety disorders and physical illness. *European Archives of Psychiatry and Clinical Neurosciences*. 2003; 253:313–320.
- Jones GT, Power C, Macfarlane GJ. Adverse events in childhood and chronic widespread pain in adult life: results from the 1958 British Birth Cohort Study. *Pain*. 2009; 143:92–96. [PubMed: 19304391]

- Juang KD, Wang SJ, Fuh JL, Lu SR, Chen YS. Association between adolescent chronic daily headache and childhood adversity: a community-based study. *Cephalalgia*. 2004; 24:54–59. [PubMed: 14687014]
- Kalin NH, Shelton SE, Barksdale CM. Opiate modulation of separation-induced distress in non-human primates. *Brain Research*. 1988; 440:285–292. [PubMed: 3359215]
- Katerndahl DA. Chest pain and its importance in patients with panic disorder: an updated literature review. *Primary Care Companion to the Journal of Clinical Psychiatry*. 2008; 10:376–383.
- Katon W. Panic disorder: epidemiology, diagnosis, and treatment in primary care. *Journal of Clinical Psychiatry*. 1986; 47(Suppl.):21–30. [PubMed: 3531189]
- Keck ME, Strohle A. Challenge studies in anxiety disorders. *Handbook of Experimental Pharmacology*. 2005:449–468. [PubMed: 16594268]
- Kendler KS, Neale MC, Kessler RC, Heath AC, Eaves LJ. Childhood parental loss and adult psychopathology in women. A twin study perspective. *Archives of General Psychiatry*. 1992; 49:109–116. [PubMed: 1550463]
- Klein DF. Delineation of two drug-responsive anxiety syndromes. *Psychopharmacologia*. 1964; 5:397–408. [PubMed: 14194683]
- Klein DF. False suffocation alarms, spontaneous panics, and related conditions. An integrative hypothesis. *Archives of General Psychiatry*. 1993; 50:306–317. [PubMed: 8466392]
- Kopce JA, Sayre EC. Stressful experiences in childhood and chronic back pain in the general population. *Clinical Journal of Pain*. 2005; 21:478–483. [PubMed: 16215332]
- Kushner MG, Beitman BD. Panic attacks without fear: an overview. *Behaviour Research and Therapy*. 1990; 28:469–479. [PubMed: 2076084]
- Lee SH, Lim J, Vannucci M, Petkova E, Preter M, Klein DF. Order-preserving dimension reduction procedure for the dominance of two mean curves with application to tidal volume curves. *Biometrics*. 2008; 64:931–939. [PubMed: 18177460]
- Liebowitz MR, Fyer AJ, Gorman JM, Dillon D, Appleby IL, Levy G, Anderson S, Levitt M, Palij M, Davies SO, Klein DF. Lactate provocation of panic attacks. I. Clinical and behavioral findings. *Archives of General Psychiatry*. 1984a; 41:764–770. [PubMed: 6742978]
- Liebowitz MR, Gorman JM, Fyer AJ, Dillon DJ, Klein DF. Effects of naloxone on patients with panic attacks. *American Journal of Psychiatry*. 1984b; 141:995–997. [PubMed: 6087689]
- Liebowitz MR, Gorman JM, Fyer AJ, Levitt M, Dillon D, Levy G, Appleby IL, Anderson S, Palij M, Davies SO, Klein DF. Lactate provocation of panic attacks. II. Biochemical and physiological findings. *Archives of General Psychiatry*. 1985; 42:709–719. [PubMed: 4015313]
- Liotti M, Brannan S, Egan G, Shade R, Madden L, Abplanalp B, Robillard R, Lancaster J, Zamarripa FE, Fox PT, Denton D. Brain responses associated with consciousness of breathlessness (air hunger). *Proceedings of the National Academy of Sciences USA*. 2001; 98:2035–2040.
- Lipsitz JD, Martin LY, Mannuzza S, Chapman TF, Liebowitz MR, Klein DF, Fyer AJ. Childhood separation anxiety disorder in patients with adult anxiety disorders. *American Journal of Psychiatry*. 1994; 151:927–929. [PubMed: 8185008]
- Liu Z, Li X, Ge X. Left too early: the effects of age at separation from parents on Chinese rural children's symptoms of anxiety and depression. *American Journal of Public Health*. 2009; 99:2049–2054. [PubMed: 19762669]
- Martinez JM, Papp LA, Coplan JD, Anderson DE, Mueller CM, Klein DF, Gorman JM. Ambulatory monitoring of respiration in anxiety. *Anxiety*. 1996; 2:296–302. [PubMed: 9160637]
- Olsson M, Ho HP, Annerbrink K, Thylefors J, Eriksson E. Respiratory responses to intravenous infusion of sodium lactate in male and female Wistar rats. *Neuropsychopharmacology*. 2002; 27:85–91. [PubMed: 12062909]
- Panksepp J. Feeling the pain of social loss. *Science*. 2003; 302:237–239. [PubMed: 14551424]
- Papp LA, Klein DF, Gorman JM. Carbon dioxide hypersensitivity, hyperventilation, and panic disorder. *American Journal of Psychiatry*. 1993; 150:1149–1157. [PubMed: 8392296]
- Papp LA, Martinez JM, Klein DF, Coplan JD, Norman RG, Cole R, de Jesus MJ, Ross D, Goetz R, Gorman JM. Respiratory psychophysiology of panic disorder: three respiratory challenges in 98 subjects. *American Journal of Psychiatry*. 1997; 154:1557–1565. [PubMed: 9356564]

- Parsons LM, Egan G, Liotti M, Brannan S, Denton D, Shade R, Robillard R, Madden L, Abplanalp B, Fox PT. Neuroimaging evidence implicating cerebellum in the experience of hypercapnia and hunger for air. *Proceedings of the National Academy of Sciences USA*. 2001; 98:2041–2046.
- Pickar D, Cohen MR, Naber D, Cohen RM. Clinical studies of the endogenous opioid system. *Biological Psychiatry*. 1982; 17:1243–1276. [PubMed: 6295518]
- Preter, M.; Klein, DF. Panic disorder and the suffocation false alarm theory: current state of knowledge and further implications for neurobiologic theory testing. In: Bellodi, L.; Perna, G., editors. *The Panic Respiration Connection*. MDM Medical Media; Milan: 1998. p. 1-24.
- Preter M, Klein DF. Panic, suffocation false alarms, separation anxiety and endogenous opioids. *Progress in Neuro-Psychopharmacology and Biological Psychiatry*. 2008; 32:603–612. [PubMed: 17765379]
- Shonkoff JP, Boyce WT, McEwen BS. Neuroscience, molecular biology, and the childhood roots of health disparities: building a new framework for health promotion and disease prevention. *Journal of the American Medical Association*. 2009; 301:2252–2259. [PubMed: 19491187]
- Sinha SS, Goetz RR, Klein DF. Physiological and behavioral effects of naloxone and lactate in normal volunteers with relevance to the pathophysiology of panic disorder. *Psychiatry Research*. 2007; 149:309–314. [PubMed: 17156855]
- Sluka KA, Deacon M, Stibal A, Strissel S, Terpstra A. Spinal blockade of opioid receptors prevents the analgesia produced by TENS in arthritic rats. *Journal of Pharmacology and Experimental Therapeutics*. 1999; 289:840–846. [PubMed: 10215661]
- Spitzer RL, Williams JB, Gibbon M, First MB. The Structured Clinical Interview for DSM-III-R (SCID). I: History, rationale, and description. *Archives of General Psychiatry*. 1992; 49:624–629. [PubMed: 1637252]
- Wallen MC, Lorman WJ, Gosciniak JL. Combined buprenorphine and chlonidine for short-term opiate detoxification: patient perspectives. *Journal of Addictive Diseases*. 2006; 25:23–31. [PubMed: 16597570]
- Weinberger SE, Steinbrook RA, Carr DB, von Gal ER, Fisher JE, Leith DE, Fencel V, Rosenblatt M. Endogenous opioids and ventilatory responses to hypercapnia in normal humans. *Journal of Applied Physiology*. 1985; 58:1415–1420. [PubMed: 3158633]

Fig. 1.

Tidal volume, respiratory rate, and minute ventilation responses in normal subjects to intravenous (i.v.) lactate preceded by naloxone (NL) *versus* i.v. lactate preceded by saline (SL): all subjects.

Fig. 2.

Tidal volume, respiratory rate, and minute ventilation responses in normal subjects to intravenous (i.v.) lactate preceded by naloxone (NL) *versus* i.v. lactate preceded by saline (SL): interaction with childhood parental loss (CPL).

Table 1
Effect of condition (NL v. SL) over the 17 min before the end of the lactate infusion^a

Effects	Tidal volume			Respiratory rate			Minute ventilation		
	Estimate	S.E.	p	Estimate	S.E.	p	Estimate	S.E.	p
Time	9.23	1.49	<0.0001	-0.02	0.03	0.54	167.35	16.88	<0.0001
Time ²	-1.06	0.34	0.0018						
NL v. SL ^b	119.50	14.57	<0.0001	-0.10	0.49	0.84	1342.1	165.34	<0.0001
(NL v. SL) × time				0.13	0.04	0.0052			
(NL v. SL) × time ²									

NL, Naloxone + lactate; SL, saline + lactate; S.E., standard error.

^a Only effects that are present in the final model are reported. The modeling begins with all effects included. Non-significant effects ($\alpha = 0.05$) are eliminated one by one following the hierarchical principle: higher-order terms are eliminated first and lower-order terms are retained in the model regardless of their significance if a higher-order effect that contains them is significant.

^b (NL v. SL) denotes an indicator for NL: NL = 1, SL = 0.

Table 2
Effect of CPL and condition (NL v. SL) over the 17 min before the end of the lactate infusion^a

Effects	Tidal volume			Respiratory rate			Minute ventilation		
	Estimate	s.e.	p	Estimate	s.e.	p	Estimate	s.e.	p
Time	9.22	1.48	<0.0001	-0.05	0.05	0.2456	218.6	23.17	<0.0001
Time ²	-1.06	0.34	0.0018						
NL v. SL ^b	157.32	20.13	<0.0001	0.04	0.67	0.9482	1800.8	227.0	<0.0001
CPL	-24.83	65.11	0.7032	0.71	1.21	0.5599	-706.8	576.1	0.2198
(NL v. SL) × time				0.22	0.07	0.0006			
(NL v. SL) × time ²									
CPL × time				0.07	0.06	0.2965	-106.8	33.44	0.0014
CPL × time ²									
(NL v. SL) × CPL	-78.80	29.06	0.0067	-0.29	0.97	0.7619	-955.6	327.64	0.0035
(NL v. SL) × CPL × time				-0.19	0.09	0.0465			
(NL v. SL) × CPL × time ²									

CPL, Childhood parental loss; NL, naloxone + lactate; SL, saline + lactate; s.e., standard error.

^a Only effects that are present in the final model are reported.

^b (NL v. SL) denotes an indicator for NL; NL = 1, SL = 0; CPL is an indicator for the presence of CPL.