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Antagonism of substance P and perception of breathlessness in patients with chronic obstructive pulmonary disease

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ABSTRACT

The objective of this study was to investigate whether substance P, an excitatory neuropeptide, modulates the perception of breathlessness by administering aprepitant, a selective antagonist that blocks neurokinin (NK)-1 receptor signaling.

Individual targeted resistive load breathing (RLB) was used to provoke breathlessness.

In Study 1, sixteen patients (age, 70 ± 6 years) with chronic obstructive pulmonary disease (COPD) reported similar ratings of breathlessness during RLB between oral aprepitant (125 mg) and placebo. After aprepitant, but not with placebo, there were significant increases in blood levels of substance P (+54 ± 39%) and beta-endorphin (+27 ± 17%). A similar design was used in Study 2 except that naloxone (10 mg) was administered intravenously prior to RLB to block any effect of endogenous opioids. Nine patients with COPD reported comparable breathlessness ratings during RLB between aprepitant and placebo.

Our results do not support a role for the substance P-NK-1 pathway in the perception of breathlessness in patients with COPD. With selective antagonism of NK-1 signaling, there was co-transmission of substance P and beta-endorphin neuropeptides.

Introduction

Excitatory and inhibitory neuropeptides orchestrate and modulate different parameters of respiratory activity by targeting specific receptors. *In vitro* testing using animal preparations have identified the effect of different neuropeptides on respiratory rhythm (Doi and Ramirez, 2008). The results of these basic science studies can be applied to investigate the modulation of dyspnea in human subjects. For example, *in vitro* studies demonstrate that endogenous opioids reduce the amplitude and frequency of breathing by acting on receptors and neurons in the pre-Boetzinger complex of the medulla (Mellen et al., 2003), whereas *in vivo* studies support their role in modulating the perception of breathlessness (Mahler et al., 2009; Bellofiore et al., 1990; Gifford et al., 2011). When naloxone was administered to block opioid receptor signaling, patients with asthma and chronic obstructive pulmonary disease (COPD) reported higher ratings of breathlessness in response to bronchoconstriction, treadmill exercise, and resistive load breathing (RLB) compared with normal saline (Bellofiore et al., 1990; Mahler et al., 2009; Gifford et al., 2011). As various endogenous neuropeptides provide continuous excitatory and inhibitory input into the respiratory network (Doi and Ramirez, 2008), the perception of dyspnea is putatively modulated in a similar manner.

Substance P is a candidate neuropeptide for modulating dyspnea, although there is no current evidence linking substance P with breathing difficulty. Along with its receptor neurokinin (NK)-1, substance P is found in the peripheral and central (CNS) nervous systems and exerts excitatory effects on respiratory rhythm, augmenting tidal volume and frequency (Bonham, 1995; Hargreaves, 2002). Various studies have demonstrated that the substance P-NK-1 receptor system has been identified in the cingulate cortex, caudate putamen, nucleus accumbens, hippocampus, amygdala, hypothalamus as well as periaqueductal gray; these regions of the brain are involved in the regulation of affective behaviors and the response to stress (Arai and Emson, 1986; Mantyh et al., 1984; Ribeiro-da-Silva and Hokfelt, 2000; Ebner and Singewald, 2006).

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Like endogenous opioids, substance P plays a key role in nociception. In response to a noxious stimulus, substance P is released from terminals of sensory nerve fibers that transmit pain impulses from peripheral NK-1 receptors to the CNS (Stein and Lang, 2009; Duggan et al., 1988; Schaible et al., 1990). Like endogenous opioids, substance P and the NK-1 receptor are present in the respiratory system, being found in bronchial smooth muscle, blood vessels, mucous glands, immune cells, and sensory nerves (Kudlacz et al., 1993).

The primary objective of this study was to investigate the role of substance P in the perception of dyspnea in patients with COPD by administering aprepitant, a selective antagonist that blocks NK-1 receptor signaling. Aprepitant is approved for the prevention of chemotherapy-induced nausea and vomiting; it crosses the blood brain barrier and alleviates the emetic effects of substance P (Curran and Robinson, 2009). Patients with the chronic bronchitis phenotype of COPD were included in the study based on the observation that levels of substance P are elevated in their induced sputum (Tomaki et al., 1995). As afferent nerves are the principal source of substance P in the airways (De Swert and Joos, 2006), we selected this COPD phenotype based on our understanding that activated airway receptors send afferent impulses to the CNS that contribute to the perception of dyspnea (Parshall et al., 2012).

The hypothesis of study 1 was that patients with COPD would report lower ratings of the intensity and/or unpleasantness of dyspnea with aprepitant compared with placebo during RLB. As blood levels of both substance P and beta (β)-endorphin increased after administration of aprepitant, study 2 was then performed to eliminate any possible role that release of endogenous opioids might affect the perception of breathlessness with NK-1 receptor blockade. In study 2, the same design was used except that 10 mg of naloxone was administered intravenously 5 min prior to RLB to block opioid receptor signaling. As these investigations were exploratory, we included three major dyspnea outcomes based on the 2012 American Thoracic Society update (Parshall et al., 2012). Both the sensory (intensity) and affective (unpleasantness) domains of dyspnea were measured along the multidimensional dyspnea profile (Meek et al., 2012).

Methods

Subjects

All subjects provided written informed consent.

Study 1. Patients were recruited from the out-patient clinic at our institution. Each patient reported breathing difficulty with activities of daily living. Inclusion criteria were: age ≥ 50 years; a diagnosis of COPD (Rabe et al., 2007); at least 10 pack-year history of smoking; a clinical diagnosis of chronic bronchitis (productive cough on most days for a minimum of three months per year for at least two successive years); post-bronchodilator forced expiratory volume in one second (FEV1) 30–80% predicted; and post-bronchodilator FEV1/FVC < 70%. Exclusion criteria were: current smoker; pregnancy; current use of narcotic medications; any concomitant disease that might interfere with study procedures; and use of a drug that may cause a possible drug interaction with aprepitant. An angiotensin converting enzyme inhibitor was not allowed as substance P is degraded by the angiotensin converting enzyme. Patients continued usual COPD medications throughout the study.

Study 2. The recruitment process as well as inclusion/exclusion criteria were identical as described for Study 1.

Study design

Both study protocols were approved by the Committee for the Protection of Human Subjects at Dartmouth College (#23276 and #23992).

Study 1. The study was a randomized, cross-over, placebo-controlled, double-blind trial with three visits (2–3 days apart). The purpose of this study was to compare the effect of aprepitant, a substance P antagonist, with a placebo on the perception of breathlessness in patients with COPD. Study medications were prepared by the investigational pharmacy at our institution.

Study 2. The study was identical to Study 1 except naloxone (10 mg) was administered intravenously prior to RLB in order to block endogenous opioid responses to aprepitant (see results of Study 1).

Interventions

Study 1. One 125 mg capsule of aprepitant or one placebo capsule containing inert powder with an identical appearance was administered orally in random order at 8:30 a.m. at Visits 2 and 3.

Study 2. One 125 mg capsule of aprepitant or one placebo capsule containing inert powder with an identical appearance was administered orally in random order at 8:30 a.m. at Visits 2 and 3. At 5 min prior to RLB, naloxone (10 mg) was administered intravenously over 1 min to block endogenous opioid responses to aprepitant (see results of Study 1) (Gifford et al., 2011).

Procedures

Study 1. The sequence of testing at all visits is described in Table 1. Levels of substance P and β-endorphin immunoreactivity were measured from venous blood by modified ELISA technique (Porstmann and Kiessig, 1992; Kraemer et al., 1989; Avrameas, 1992).

Study 2. The sequence of testing at all visits is similar as described in Table 1 except that blood levels of substance P and β-endorphin immunoreactivity were not measured. As noted above, naloxone (10 mg) was administered intravenously 5 min prior to RLB.

Statistical analysis

Ratings of the intensity and unpleasantness of breathlessness for each patient during RLB were primary outcomes (Gifford et al., 2011). Time-weighted averages of ratings of breathlessness were compared using the Mann–Whitney test [MedCalc for Windows, version 12.7.7 (MedCalc Software; Ostend, Belgium)]. As patients did not provide ratings of breathlessness before RLB, no pre-post RLB comparisons were made. Paired-*t* tests were used to compare immediate sensory intensity, immediate unpleasantness, and emotional response scores on the multidimensional dyspnea profile (Meek et al., 2012) after RLB between aprepitant and placebo. A repeated measures analysis of variance with Bonferroni correction for multiple comparisons was used to test for differences in neuropeptide blood levels with each intervention (GraphPad Prism version 6.03 for Windows, GraphPad Software, La Jolla, California, USA). Spearman correlation analysis was performed to examine the relationship between changes in blood levels of substance P and β-endorphin at different time periods. A p value < 0.05 was considered statistically significant. For Study 1, a sample size of 16 was adequate to provide 80% power to detect a significant difference in breathlessness ratings (alpha = 0.05) (Mahler et al., 2009; Pepin et al., 2005).