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Interaction of noscapine with the bradykinin mediation of the cough response

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Angiotensin Converting Enzyme Inhibitors (ACEI) like captopril and enalapril, can induce persistant cough in man. Noscapine, an antitussive alkaloid, can be used to suppress ACEI-induced cough. Some workers have suggested a role for bradykinin in precipitation of ACE-induced cough. Work carried out in our laboratory has shown noscapine to be a non-competitive inhibitor of bradykinin in guinea pig ileum. It is therefore possible that noscapine suppresses cough by blocking the effect of bradykinin receptor activation in the airways.

Guinea pigs were placed in a cough-chamber connected to an air pump and a pressure transducer. Capsaicin was sprayed into the chamber and cough was recorded as a distinctive change in air pressure inside the cough-chamber.

Animals treated with 1 mg/kg captopril and enalapril for 7 days, showed increased cough response. Ten μg/kg FR190997, a non-peptide agonist of the bradykinin B2 receptor, also increased the cough response. Noscapine at 0.5, 1 and 2 mg/kg was able to reverse the effects of ACEI and FR190997. Naloxone, a specific opioid receptor inhibitor, did not block the antitussive effects of noscapine in enalapril or FR190997 treated guinea pigs. This antitussive effect of noscapine is not mediated via the μ, κ or δ opioid receptors. It is therefore possible that noscapine exerts its antitussive action by interfering with the bradykinin cough mediation.

Keywords: cough, noscapine, bradykinin, guinea pig

Noscapine is an isoquinoline alkaloid found in opium (5). Unlike most other alkaloids obtained from the opium latex, this drug is devoid of any significant analgesic, sedative or euphoriant effect generally associated with this group of drugs. Although recently, this drug was shown to be an inducer of apoptosis in some cell lines (9), the only important clinical effect associated with noscapine is its antitussive activity (5, 9).

There are a number of endogenous mediators involved in the cough reflex, these include bradykinin (3), histamine (11), substance P (7) and some prostaglandins (11). Of these, bradykinin and substance P have been studied most widely. It is now

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understood that both these mediators can have a potentiating effect on the cough reflex. Increased release of Substance P leads to a neurogenic inflammation of the airways (3). Bradykinin on the other hand can cause: 1. Increased release of substance P; 2. Release of histamine from mast cells (via B2 receptor activation); 3. Increased synthesis of prostaglandins (3). The overall effect that bradykinin and substance P exert, appears to be increased sensitivity of the airway sensory nerves, leading to increased sensory input in the putative "cough centers" in the CNS. This in turn, causes easier initiation of the motor components of the cough reflex.

Opioid antitussive drugs can suppress cough (2) by acting on either the opioid receptors on the airway sensory nerves or the opioid receptors located in the CNS. Agonists of the μ, σ and δ opioid receptor are believed to have central antitussive effects (6, 8).

It is possible that the antitussive effects of noscapine are due to the interaction of this drug with one or more of these mediators. Recent work in our laboratory on the contractile effects of bradykinin and FR190997, a synthetic bradykinin receptor agonist, on the guinea pig ileum has shown noscapine to be a non-competitve inhibitor of these compounds (8). It is possible that the antitussive effects of noscapine are also due to its interfering with the action of bradykinin.

Materials and Methods

Noscapine, naloxone hydrochloride and codeine were gifts from Temad Pharmaceutical Company, Tehran, Iran. Capsaicin, captopril and enalapril were obtained from Sigma, Illinois, USA. FR190997 was a kind gift from Fujisawa Pharmaceutical Company, Osaka, Japan.

Male Dunkin–Hartley guinea pigs (350–450 g) were placed in a clear plastic exposure chamber with an internal volume of 2 litres. Air was pumped into the chamber at a rate of 2 l/min during the experiment. The internal air-pressure changes of the chamber were recorded using a pressure transducer mounted on the top surface of the chamber, connected to a Physioscribe recorder (Stoelting Co., Chicago, Illinois, USA). Cough was recorded as a greater than normal inspiration followed by a rapid expiration. Animals were exposed to an aerosole of cough inducing compound and air-flow changes were recorded for a period of 5 min. A 10 mg/ml Noscapine stock solution was prepared in 0.1 M acetic acid and diluted with saline prior to injection. Codeine, naloxone hydrochloride, enalapril, and captopril solutions were prepared in saline. One ml drug solution was injected i.p. 1 h before the cough challenge, and when appropriate 1 ml vehicle was injected i.p. as control. Seven animals were used for each experiment.

SPSS and MS Excel software packages were used to carry out ANOVA and Student's *t*-test on the data.

Results

When guinea pigs were exposed to nebuilised capsaicin, a respiratory irritant, a characteristic pattern of air pressure change occurred. This pattern of air pressure change was induced by capsaicin, dose dependently, over a range of 1.6×10^{-6} M to 1.6×10^{-4} M (Fig. 1). To ensure that capsaicin induced changes in exposure chamber air pressure were in fact due to cough induction and not sneezing, the animals were pretreated with codeine, an antitussive drug. Codeine treated animals showed a significantly ($p<0.01$) decreased response (2.82±0.72 coughs/5 min) compared to the control group (9.25±0.24 coughs/5 min). A concentration of 1.6×10^{-4} M capsaicin was used to induce cough in all subsequent experiments.

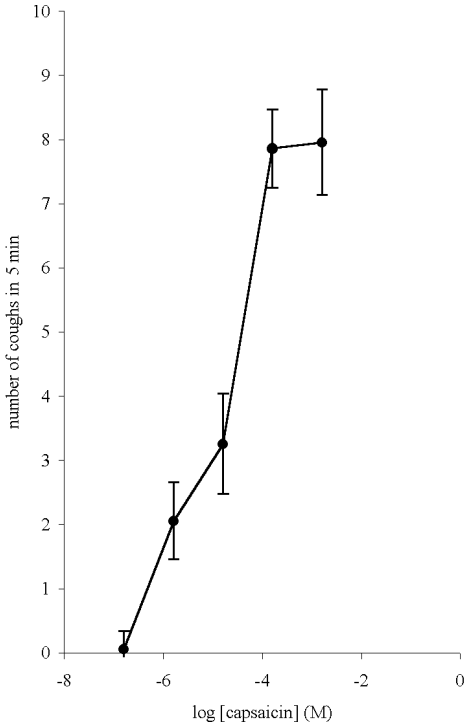

Fig. 1. Effects of different concentrations of capsaicin on cough induction. Guinea pigs were exposed to nebulised solutions of capsaicin in ethanol-saline (50:50) for 5 min and cough response was recorded. The cough response was corrected by subtraction of the response caused by the vehicle. The graph shows the mean number of coughs ± SD, n=7

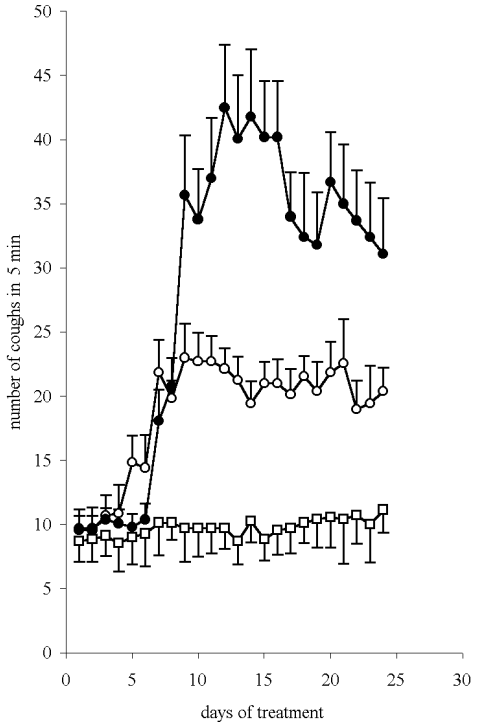

Fig. 2. Effects of long-term treatment with enalapril and captopril on capsaicin induced cough in guinea pigs. Guinea pigs were injected with 1 mg/kg enalapril (●), 1 mg/kg captopril (○) or saline (□) intra ± SD, n=7

Long-term treatment with both enalapril and captopril increased capsaicin induced cough in guinea pigs (Fig. 2). Captopril induced increase in the cough response was statistically significant from day 4 to day 6 ($p<0.01$) and from day 7 to day 24 ($p<0.001$). Enalapril induced increase in the cough response was statistically significant ($p<0.001$) from day 7 onwards. After day 9, enalapril caused a much greater increase in the cough response compared to captopril ($p<0.001$). As enalapril was more effective in increasing the capsaicin induced cough response, in all subsequent experiments the animals were pretreated for 7 days with 1 mg/kg i.p. enalapril.

Noscapine reduced enalapril-increased cough response when used at 0.5, 1 and 2 mg/kg doses ($p<0.001$) (Fig. 3). Naloxone at 3 and 6 μg/kg i.p. was unable to reverse the cough suppressive effects of noscapine (Fig. 4) while at 6 μg/kg, it significantly decreased ($p<0.001$) the antitussive effects of codeine (Fig. 5).

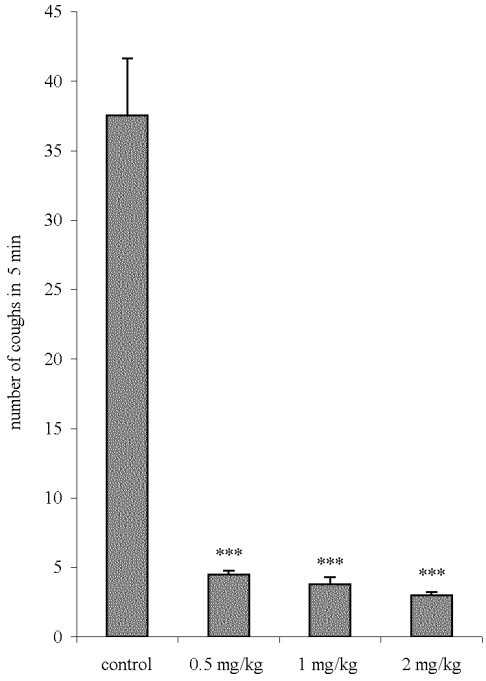

Fig. 3. Effect of different doses of noscapine on enalapril increased cough response. Guinea pigs were treated with 1 mg/kg enalapril i.p. for 7 days. Noscapine was injected i.p. 1 h before exposure to capsaicin. The graph shows the mean number of coughs ± SD, n=7 (***$p<0.001$ for all groups compared to control group)

FR190997, a nonpeptide agonist of the bradykinin receptor, at a dose of 10 μg/kg increased capsaicin induced cough in guinea pigs ($p<0.05$). Noscapine at 1 mg/kg was able to decrease the tussive effects of FR190997 significantly ($p<0.001$) (Fig. 6).

Discussion

Angiotensin Converting Enzyme (ACE) Inhibitors are used for the treatment of hypertension and heart failure. One of the irritating side-effects of these drugs is a dry, nonproductive cough which is frequently reported in patients. Although the mechanism of this tussive action is not entirely understood, decreased degredation of bradykinin may be involved (4). Noscapine used clinically as an antitussive drug can decrease the cough induced by ACEI's in patients, but its mode of action is not completely known.

Work carried by Mahmoudian et al. (10), suggests that in the guinea pig ileum, noscapine acts as a noncompetitive antagonist of bradykinin. This work was carried out in order to determine whether noscapine exerted its antitussive effects by interacting with the bradykinin mediation of cough.

Awake guinea pigs were selected as the animal model for cough as it appears that they produce the most consistent response to inhaled irritants (8, 13). 1.6×10^{-4} M capsaicin produced maximal effect and was used in all experiments to induce cough (Fig. 1).

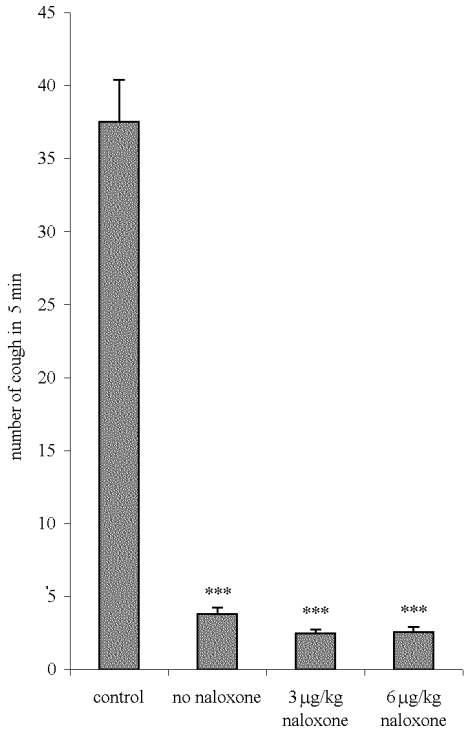

Fig. 4. Effects of naloxone on cough suppressive effects of noscapine. Guinea pigs were treated with 1 mg/kg enalapril i.p. for 7 days. Noscapine (1 mg/kg) and naloxone were injected i.p. 1 h before exposure to capsaicin. The graph shows the mean number of coughs ± SD, n=7 (***p<0.001 for all groups compared to control group)

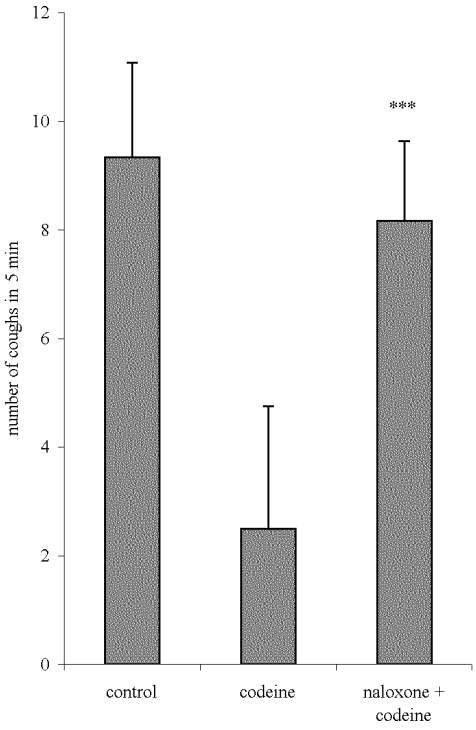

Fig. 5. Effects of naloxone on cough suppressive effects of codeine. Guinea pigs were administered 1 mg/kg noscapine and 1 mg/kg codeine i.p. 1 h before exposure to capsaicin. The graph shows the mean number of coughs ± SD, n=7 (***$p<0.001$ for codeine + naloxone group compared to codeine group)

Guinea pigs treated with enalapril and captopril, every day for 24 days, showed delayed increased sensitivity to capsaicin. Increased cough response in captopril-treated animals became significant after 5 days and reached maximum by day 8. In enalapril treated group, increased cough response was observed on day 7 which reached a maximum on day 9. Enalapril was the more effective drug in increasing the cough response (Fig. 2). This corresponds with data obtained by other workers (3, 4, 12). The maximal cough inducing effect of enalapril, reached on day 9, caused the animals to cough approximately once every 6 seconds. In order to cause less suffering to the animals, it was decided that the animals would be pretreated for 7 days with enalapril for all subsequent experiments.

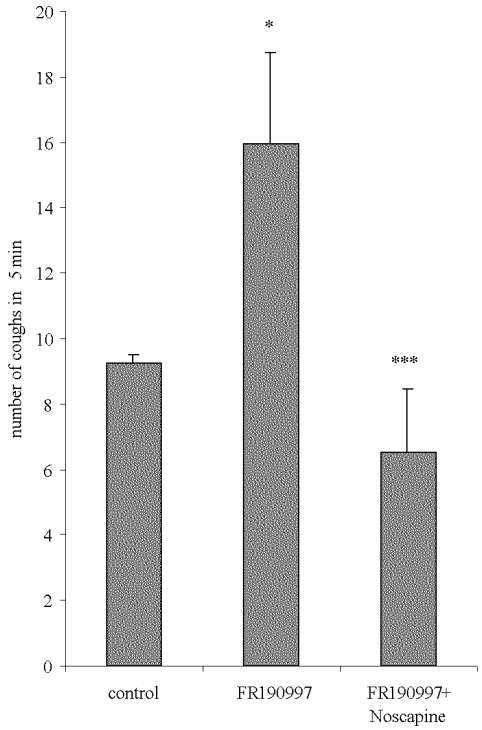

Fig. 6. Effect of noscapine on increase in cough induction by FR190997. Guinea pigs were administered 10 μg/kg FR190997 with or without 1 mg/kg noscapine 1 h before exposure to capsaicin. The graph shows the mean number of coughs ± SD, n=7 (*$p<0.05$ for FR190997 group compared to control group and ***$p<0.001$ for FR190997 group compared to FR190997 + Noscapine group)

Administration of noscapine to guinea pigs treated with enalapril, decreased the cough response (Fig. 3). Three and 6 μg/kg naloxone failed to reverse the effects of noscapine (Fig. 4). Codeine at 1 mg/kg inhibited capsaicin induced cough, this effect was blocked by 6 μg/kg naloxone (Fig. 5). Naloxone has an antagonistic action at the μ, κ and to a lesser extent the δ opioid receptor. Naloxone could not reverse the anti-tussive effect of noscapine, therefore, it is unlikely that the antitussive effects of noscapine are mediated through the μ, κ or δ opioid receptors. Codeine, on the other hand, exhibited an antitussive effect which was clearly reversed by naloxone. Therefore, codeine does appear to act via a subtype of the opioid receptor. Thus, these two antitussive drugs, seem to suppress cough via two different mechanisms.

FR190997, is a synthetic nonpeptide bradykinin agonist with high affinity for the B2 bradykinin receptors (1). Administration of FR190997 to guinea pigs caused an increase in the responsiveness of the animals to the cough challenge. This tussive effect too was blocked by noscapine (Fig. 6).

In conclusion, increased bradykinin receptor stimulation, whether caused by ACEI administration or synthetic bradykinin agonist FR190997, stimulates the cough response, probably through B2 receptors. Noscapine, a nonnarcotic antitussive drug, inhibits the tussive effects of bradykinin receptor activation. This antitussive effect of noscapine is not mediated via μ, κ or δ opioid receptors. It is therefore possible that noscapine exerts its antitussive action by interfering with the bradykinin cough mediation.

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