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Characteristics of prostaglandin induced cough in man

J. F. COSTELLO, L. S. DUNLOP & P. J. GARDINER[1]
Chest Unit, Kings's College Medical School, Denmark Hill, London and [1]Research Department, Miles Laboratories Ltd, Slough, Bucks

1 Inhaled $PGF_{2\alpha}$ and PGE_2 were evaluated for relative tussive activity to non-prostanoid tussive agents. In addition a comparison was sought between the present observations and those in the cat, the only laboratory animal which consistently coughs to prostanoids.
2 Five healthy volunteers were repeatedly challenged at 90 min intervals with aerosols of PGE_2 (100–500 $\mu g\ ml^{-1}$) and tussive activity was monitored. In a second separate study again monitoring tussive activity 10 healthy volunteers inhaled aerosols of either $PGF_{2\alpha}$ (0.1–100 $\mu g\ ml^{-1}$), PGE_2 (0.1–100 $\mu g\ ml^{-1}$), acetylcholine (0.1–50 $mg\ ml^{-1}$) or citric acid (5–20% w/v) in a randomised procedure.
3 Objective measurement of tussive activity was achieved using a throat microphone linked via a discriminator to a pen recorder.
4 All four compounds produced two distinct phases of tussive activity, an early phase during challenge and a late phase 1–15 min post-challenge.
5 Repeated challenges with PGE_2 produced significant ($P < 0.01$) tachyphylaxis to the late phase responses only.
6 Both $PGF_{2\alpha}$ and PGE_2 were approximately 1000 and 10,000 times more potent than acetylcholine and citric acid respectively for both phases of tussive activity.
7 Tussive activity was accompanied with retrosternal soreness and tightness of the chest for PGE_2, increased sputum for $PGF_{2\alpha}$, and sore throats with citric acid.
8 Although a correlation exists for man and cat with regards to the tussive potency and the early and late phases of $PGF_{2\alpha}$ activity no such correlation seems to exist for PGE_2. The high tussive potency of the prostaglandins in man suggest that their local release in various respiratory pathophysiological conditions may be responsible for the accompanying coughs/irritancy.

Keywords tussives $PGF_{2\alpha}$ PGE_2 acetylcholine citric acid

Introduction

It has been known for many years that inhaled prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$), PGE_2 or PGE_1 produce coughing, retrosternal soreness and significant changes in airway tone in normal and asthmatic volunteers. $PGF_{2\alpha}$, and to a lesser extent PGE_2, generally induce bronchoconstriction in man whereas PGE_1 predominantly elicits a bronchodilator response (Cuthbert, 1969; Kawakami *et al.*, 1973; Mathe & Hedqvist, 1975).

The unwanted irritant effects of PGE_1 and PGE_2 have probably been the major factors limiting their therapeutic use as bronchodilators.

Correspondence: Dr J. F. Costello, Chest Unit, King's, College Medical School, Denmark Hill, London

Many synthetic derivatives of PGE_2 have been prepared in an attempt to overcome this problem. Unfortunately such compounds were prepared and tested before a full understanding of prostaglandin activity was attained. Consequently due to the poor bronchodilator selectivity of PGE_2 and its analogues generally all of the candidates produced coughing and/or bronchoconstriction and failed to progress further (Grudzinskas *et al.*, 1980).

More recently synthetic derivatives of PGE_1 were developed in an attempt to increase bronchodilator selectivity at the expense of tracheobronchial irritancy and bronchoconstriction. Such compounds were evaluated for their tussive effects using the cat cough/irritancy test (Gardiner & Collier, 1980). Surprisingly following numerous studies with a range of laboratory animals the cat was shown to be virtually the only species sensitive to the tussive activity of inhaled prostaglandins. Using this test model it was proposed that TR4 161, a carbinol derivative of PGE_1 would be a useful new bronchodilator with little or no irritant activity in man. However, subsequent clinical evaluation of this analogue by aerosol administration, in two studies, which demonstrated consistent bronchodilator activity, showed that it induced coughing and retrosternal soreness, albeit less than PGE_2 (Nizankowska *et al.*, 1985). Naturally such results led us and others to question the value of the feline cough/irritancy test system in predicting such activity in man.

We decided to investigate the nature of prostaglandin-induced cough in man in an attempt to determine whether such effects correlate with those reported in cats (Gardiner *et al.*, 1978; Gardiner & Browne, 1984). The study was conducted in two stages, the first to ascertain whether or not repeated inhalations of a prostaglandin elicited tachyphylaxis as had been observed with cats, and the second to determine the relative tussive potency of prostaglandins to reference non-prostaglandin tussive agonists.

Methods

Tachyphylaxis to PGE_2

Five non-smoking volunteers with no history of asthma or recent respiratory tract infection were studied. All subjects gave full informed written consent. Approval for the study was given by the Ethics Committee, King's College Hospital. PGE_2 was administered using a Wrights nebuliser driven by compressed air at a flow rate of 7 l min^{-1}. Subjects were instructed to take normal hard breaths for 1 min. Coughing was monitored using a throat microphone during inhalation and for the subsequent 15 min. The number of coughs was recorded on a Devices recorder via a discriminator which consisted of an amplifier and an R.C. network whereby the output was directly proportional to the rate of change of the input signal. An approximate indication of cough intensity could be obtained by measuring the duration of the recorded signal. Any subjective comments were also noted.

On the first day of the study subjects were given an inhalation challenge with a solution of PGE_2 (1 $\mu g\ ml^{-1}$), and coughing was monitored. No previous inhalation of any drugs had been performed with these subjects before this study day. If the subject failed to cough 5–10 times during the challenge then it was repeated using a stock solution of higher concentration until the required level of activity was achieved. The subject was then challenged with this solution at 90 min intervals for the next 6 h, i.e. a total of four challenges using the irritant solution. The results were plotted as a histogram of mean number of coughs ± 1 s.e. mean against (a) 0–1 min challenge time and (b) 1–15 min post-challenge.

Dose-response curves

Ten non-smoking volunteers with no history of asthma or recent respiratory tract infection were studied. All subjects gave their informed consent and approval for the study was given by the Ethics Committee, King's College Hospital. All agents were administered using the Wrights nebuliser driven by compressed air at a flow rate of 7 l min^{-1}. The agents used were PGE_2, 0.1, 1.0, 10.0, 100.0 $\mu g\ ml^{-1}$; $PGF_{2\alpha}$ 0.1, 1.0, 10.0, 100.0 $\mu g\ ml^{-1}$; acetylcholine chloride 0.1, 1.0 10.0, 50.0 mg ml^{-1} and citric acid 5, 10 and 20% (w/v) solutions. Each subject was challenged with these solutions using a randomised procedure on separate days, with at least three days between each inhalation to avoid the development of tachyphylaxis. Coughing was monitored and classified as previously described and dose-response curves plotted.

Materials

PGE_2 (Dinoprostone) and $PGF_{2\alpha}$ (Dinoprist) Upjohn Ltd.
Acetylcholine, BDH Biochemicals.
Citric acid, Fisher Scientific Company.

All drugs were prepared as stock solutions in phosphate buffered saline (PBS) pH 7.2. Dilutions were also prepared in this PBS, consequently

the aerosol stock solutions of $PGF_{2\alpha}$, PGE_2 and acetylcholine all had a similar pH range of 6.6–7.2. The highest aerosol stock solution of citric acid however was acidic (pH 3.0) although subsequent dilutions were within the range 4–6.8.

Results

During preliminary experiments with the prostaglandins it was noticed that no significant systemic changes occurred; however, two distinct phases of coughing were observed. An acute early phase during aerosol challenge was seen which immediately decreased or ceased on stopping challenge. Following a variable interval of 1–5 min, however, all subjects had a second phase of coughing which generally lasted 10–15 min. Consequently all results were classified as early (during challenge 0–1 min) or late (1–15 min after challenge) responses. On no occasion did the early phase coughs completely or partially interrupt aerosol inhalation.

Tachyphylaxis to PGE_2

PGE_2 induced significant early irritant activity at 100 μg ml^{-1}, 250 μg ml^{-1}, and 500 μg ml^{-1} for 3, 1, and 1 volunteers respectively. The results are expressed in Figure 1. Repeated challenges produced little or no tachyphylaxis to the early phase coughing; however, there was a significant ($P < 0.01$ by paired Student's *t*-test analysis) reduction in the number of coughs elicited in the late phase. The final challenge induced less than half the original irritant activity in the late phase response. Therefore, all further challenges were spaced at a minimum of 3 day intervals as a precautionary measure against tachyphylaxis.

Dose-response curves

Examination of the early phase responses shows that $PGF_{2\alpha}$ and PGE_2 produced similar effects with their dose-response curves plateauing at 1 μg ml^{-1} and then rising again at 10 μg ml^{-1}, although this latter effect was only significant for PGE_2 (Figure 2). Such a biphasic dose-response curve was not obtained with acetylcholine or citric acid. Although the two latter compounds were much less potent tussive agents than the prostaglandins, they produced very steep dose-response curves and over the dose range studied had comparable if not superior maximal efficacies to the prostaglandins for the early phase responses.

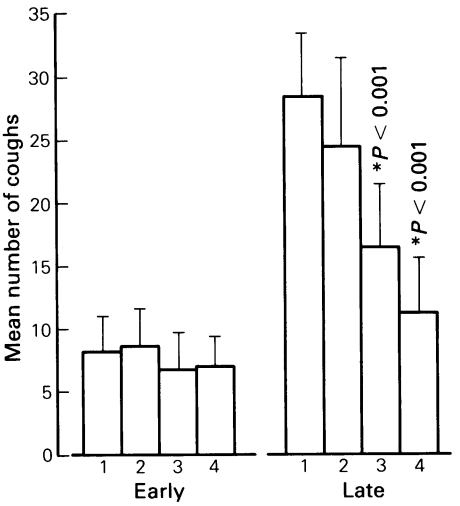

Figure 1 Tussive activity of repeated inhalation challenges with PGE_2. The concentrations of aerosol stock solution producing 5–10 coughs in these five volunteers were 100, 250 and 500 μg ml^{-1}. 'Early' and 'late' are the stages of coughing monitored during challenge, 0–1 min and 1–15 min post challenge respectively. 1, 2, 3 and 4 represent four separate successive challenges given at 90 min intervals. Each column is the mean of observations from five healthy volunteers over a 1 min period for early responses and over a 14 min period for late responses. The vertical bars are ± 1 s.e. mean and Student's paired *t*-test was used to determine whether the early or late phase response differed significantly from the first challenge response.

In contrast to the biphasic dose-response curve produced by PGE_2 and possibly $PGF_{2\alpha}$ during the early coughing phase, the late phase reponses were single probably sigmoidal dose-response curves although maximal responses could not be achieved as we considered this a safety hazard. $PGF_{2\alpha}$ was both more potent and effective than PGE_2 over the dose range studied (Figure 3). Acetylcholine and citric acid had lower maximal efficacies than $PGF_{2\alpha}$ in the late phase response and they were approximately, 1000 and 10,000 times less potent respectively than this prostaglandin for both phases of coughing. No significant difference seemed to occur in the intensity of coughing induced by all four agents.

Five subjects complained of retrosternal soreness and a tight chest following inhalation of PGE_2 (100 μg ml^{-1}). Two subjects noted increased sputum production following inhalation of $PGF_{2\alpha}$ (100 μg ml). All subjects commented on the sharp taste of citric acid, and two complained of a sore throat following inhalation of the 20% solution.

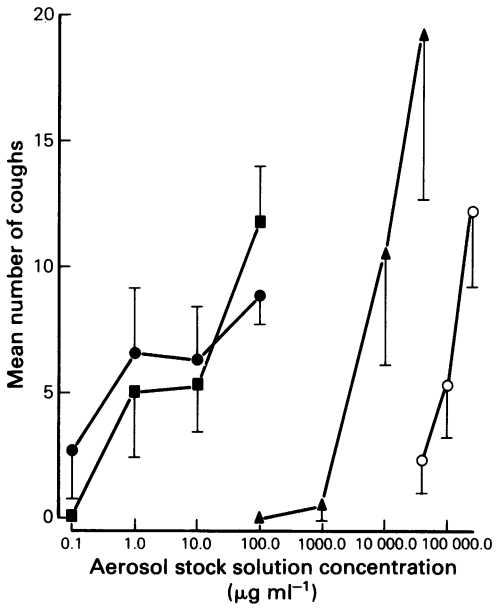

Figure 2 Early phase activity of tussive agents. Ten healthy volunteers were separately challenged with aerosols of $PGF_{2\alpha}$ (●), PGE_2 (■) acetylcholine (▲) or citric acid (○). Each point is the mean of ten observations monitored during the 1 min aerosol challenge and the vertical lines represent ± 1 s.e. mean.

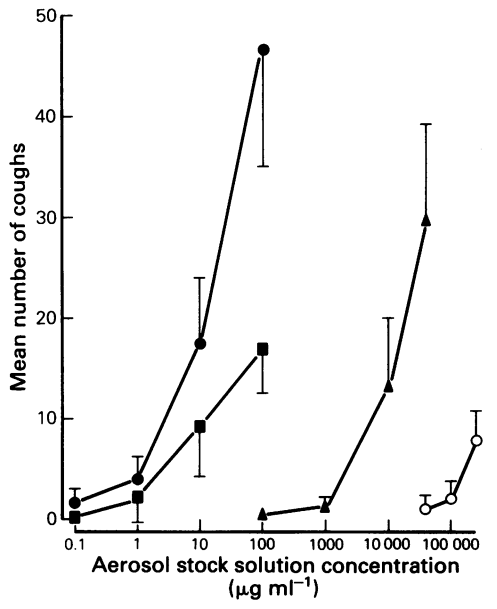

Figure 3 Late phase activity of tussive agents. Ten healthy volunteers were challenged with either $PGF_{2\alpha}$ (●), PGE_2 (■) acetylcholine (▲) or citric acid (○). Each point represents the mean observations 1–15 min after challenge and the vertical lines are ± 1 s.e. mean.

Discussion

The present study demonstrates that $PGF_{2\alpha}$ and PGE_2 have comparable maximal efficacies as tussive agents and they are markedly more potent than the established tussive agents acetylcholine or citric acid. It was not surprising that tachyphylaxis developed to repeated challenges of PGE_2 as this was reported in an earlier study by Herxheimer & Roetscher (1971). It was surprising, however, to find that two distinct phases of irritancy seemed to exist and that tachyphylaxis only developed to the second phase. Whether or not the latter phenomenon occurs with the other three agents remains to be determined. As to the two phases of tussive activity we speculate that the initial phase involves the stimulation of irritant receptors and/or local reflex pathways. The secondary phase may represent increased mucus secretion and subsequent irritancy during its clearance and/or bronchoconstriction leading indirectly to tussive activity. Increased mucus secretion may also be responsible for the biphasic shape of the early phase dose-response curve to PGE_2 and perhaps $PGF_{2\alpha}$.

The results reported here also indicate that there are some similarities in the nature of cough induced by PGs in man and cat. Both species exhibit tachyphylaxis to PGE_2 and $PGF_{2\alpha}$ is significantly more potent in both than is acetylcholine or citric acid. However, in contrast, with the observations of Gardiner *et al.* (1978) and Gardiner & Browne (1984) in the cat the early phase activity of PGE_2 in our human studies closely resembled $PGF_{2\alpha}$ with regards to potency, efficacy and the biphasic shape of the dose-response curve, such that man appears to be considerably more sensitive to PGE_2 than cats. Furthermore, PGE_2 in man produced a significant degree of late phase coughing which was not observed in the cat studies. Overall therefore it would seem that the cat cough test is a poorly predictive index of PGE_2 irritancy in man. This may be due to a difference in irritant receptor densities or to a variation in the involvement of the reflex or mucosecretory effects of PGE_2 in the two species. It may be that the PG irritant threshold dose level in the cat is higher than that of man. This may explain why the PGE_1 analogue TR4161 which was shown to be inactive in the cat cough test had moderate tussive activity in man.

A more speculative but nevertheless noteworthy point arises from the present study in the possible involvement of arachidonate cyclooxygenase metabolites (PGs and TXs), in physio-

logical and/or pathophysiological cough whether it be associated with asthma or other pulmonary pathophysiological states. Although the concentrations of stock solutions used to generate prostaglandins in the present study seem high, the doses actually reaching the lungs and thus making contact with irritant receptors, smooth muscle or mucoserous cells/glands are probably very small. Such levels may be similar to that expected in pathosphysiological states involving mast cell degranulation and bronchial smooth muscle contraction in which large amounts of PGs may be released (Lewis *et al.*, 1982; Cuthbert & Gardiner, 1983). It is possible that one consequence of this PG release may be coughing as is frequently experienced by many asthmatics. Similarly, in respiratory tract infections which involve a significant inflammatory component, generation of arachidonate metabolites is almost certain to occur and these could possibly initiate coughing and general tracheobronchial discomfort.

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