The importance of bradykinin and histamine in the skin response to antigen

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On three separate occasions 12 atopic subjects were injected intradermally with two doses of antigen and one of saline as control. Pretreatment with terfenadine 60 mg orally significantly inhibited the flare response to both the lower dose of antigen and to saline ($P < 0.05$). Ingestion of enalapril 5 mg orally 3 h before increased the flare response to both doses of antigen. Neither enalapril nor terfenadine affected the weal response when compared with placebo. Both endogenous histamine and bradykinin appear to be released during the intradermal flare response but are not important in the weal reaction to antigen.

Keywords histamine bradykinin skin enalapril terfenadine

Introduction

When exogenous histamine is injected intradermally the weal, flare and itch resemble the initial reaction to injected antigen in atopic individuals. However, it cannot be concluded that histamine is the predominant mediator released by antigen stimulated degranulation of mast cells since many other cells and mediators are involved. Although anti-histamine drugs effectively block the weal and flare of skin challenge with histamine (Crossman & Fuller, 1988), their effect on the response to antigen is less marked; some preparations such as chlorpheniramine are ineffective even when injected locally at the same site as the antigen (Phillips *et al.*, 1983).

Enalapril inhibits kininase II, one of the enzymes which degrades bradykinin, and 5 mg given orally significantly potentiates the weal response to intradermal bradykinin in normal subjects (Fuller *et al.*, 1987). Bradykinin is produced during allergic reactions (Proud *et al.*, 1983) and when injected intradermally causes a marked weal (Basran *et al.*, 1982). Therefore, bradykinin could mediate the weal response to antigen challenge and if released, should be potentiated by enalapril.

To determine the relative importance of histamine and bradykinin in the allergic reaction to injected antigen, two doses of antigen were given intradermally to atopic volunteers after oral placebo, enalapril or terfenadine.

Methods

Twelve atopic individuals who gave informed consent were studied. There were eight males and four females aged 24–40 years and they weighed 58–90 kg. The study was approved by the hospital Ethics Committee. For the pre-study assessment, increasing doses of house dust mite or grass pollen antigen (Pharmacia, Sweden) were given as 50 μ l intradermal injections with a 27 gauge needle. The antigen solutions were diluted with 0.9% saline to give concentrations of 0.5, 1, 5, 10, 50 and 100 units ml^{-1} . A positive test of at least a 2 cm diameter flare reaction to intradermal antigen was necessary for inclusion in the study and this dose of antigen, together with a dose one tenth the concentration, and a saline control were used to produce a dose-response.

For the study the volunteers took either placebo, enalapril 5 mg or terfenadine 60 mg orally in randomized order on three occasions 1 week apart. Three hours after tablet ingestion, saline and two doses of antigen were injected as in the preliminary study. Injections were into the skin of the back for men and flexor aspect of the forearm for women. For each subject the same areas of skin were used on the 3 study days.

The flare was measured 15 min after injection by tracing the area onto a perspex sheet. At 20 min after injection the perimeter of the weal was traced with ink and the ink removed by adhesive tape. The areas of weal and flare were later determined by planimetry. The traces were drawn by an independent observer who was blind to the protocol.

Results

Seven subjects received grass pollen and five house dust mite antigen. Figures 1 and 2 show the effect of placebo, terfenadine and enalapril on the weal and flare response. Compared with placebo, terfenadine significantly inhibited the flare response to saline and the low dose of antigen ($P < 0.05$). Enalapril significantly increased the flare response to both doses of antigen ($P < 0.05$) when compared with placebo. The weal response was not significantly affected by either drug when compared with placebo. Significance was tested by two tailed paired Student's *t*-test.

Discussion

Histamine is released during intradermal antigen challenge (Heavey *et al.*, 1984) but its importance is not clear. Of the many other potential mediators, kinins are likely to play a significant role (Lewis, 1970) and are known to be released during allergic reactions (Proud *et al.*, 1983).

Enalapril 5 mg orally increases the weal caused by exogenous bradykinin (Fuller *et al.*, 1987) and this is by the inhibition of kininase II, one of the enzymes which degrade bradykinin. Bradykinin may be synthesized by normal skin (Regoli & Barabe, 1980) and if released during antigen challenge would be potentiated by enalapril.

The effect of enalapril on the flare response, with no effect on weal, following intradermal antigen was unexpected. Flare involves an axon reflex in sensory nerves, although the neurotransmitter released from the afferent nerves has not been identified (Barnes *et al.*, 1986). An

Figure 1 Effect of placebo (P), terfenadine 60 mg orally (T) and enalapril 5 mg orally (E) on the weal response to intradermal saline, first dose of antigen (Ag) and a tenfold dose of antigen (Ag $\times$ 10). Data are mean + s.e. mean for 12 subjects.

Figure 2 Effect of placebo (P), terfenadine 60 mg orally (T) and enalapril 5 mg orally (E) on the flare response to intradermal saline, first dose of antigen (Ag) and a tenfold dose of antigen (Ag $\times$ 10). Data are mean + s.e. mean for 12 subjects. * = $P < 0.05$, compared with placebo.

increased flare suggests an interaction with a receptor on the sensory nerves that facilitate the axon reflex. However, kinins cause a weal rather than a flare when injected intradermally (Basran *et al.*, 1982). Others mediators that are substrates for kininase II may be involved, but we have no hypothesis as to their identity.

Antihistamine drugs can diminish the response to skin antigen challenge; a lack of specificity of the drugs used may explain some of the variability observed with cyproheptadine (Johnson *et al.*, 1984), ketotifen (Phillips *et al.*, 1983) and dimethindene maleate (Johnson *et al.*, 1984).

Terfenadine 60 mg orally is an effective inhibitor of the intradermal response to histamine, but not bradykinin (Crossman & Fuller, 1988). Studies have shown attenuation of antigen challenge by terfenadine on skin prick tests. This effect was most marked in a study of children (Kjellman & Andersson, 1986), but the maximum weal area observed was only 24 mm², which is difficult to assess. Howarth & Holgate (1984) reported terfenadine diminished weal area by an average of 11%, although the same

group later found a single dose of terfenadine or three doses of astemizole caused approximately a 50% reduction in weal diameter (approximately a 75% reduction in area) in response to skin prick testing with antigen (Holgate, 1987).

Skin responses were measured in the present study at 15 and 20 min after injection since preliminary observations indicated that this is the peak response time. Other studies examined earlier time points and if histamine is more important in the first 10 min of the reaction, then this may explain why terfenadine had no effect on weal in the present study.

The present data, taken in conjunction with the observed poor response of hay fever (Howarth & Holgate, 1984), asthma (White & Eiser, 1983) and urticaria (Mosko & Marshall, 1954) to anti-histamine drugs, suggest that histamine is not the main mediator released by antigen challenge, although it may significantly enhance other mediators. Enalapril increased the flare response to antigen probably by potentiating the effect of bradykinin released during such a reaction.

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