Immunology 1979 **38** 385

The respiratory response of sensitized rats to challenge with antigen aerosols*

HANNA PIECHUTA, M. E. SMITH, N. N. SHARE & G. HOLME *Pharmacology Department, Merck Frosst Laboratories, Kirkland, Quebec, Canada*

Accepted for publication 10 May 1979

Summary. An experimental procedure was developed to evaluate the effects of challenging sensitized conscious rats with antigen aerosols. The challenge resulted in changes in the respiratory patterns which were antigen specific and mediated by IgE antibodies. The response was inducible in non-sensitized rats by passive administration of IgE-rich serum. Sprague–Dawley rats were heterogeneous with respect to the respiratory response. A proportion (20–70%) of each group had continuous dyspnoea and other symptoms similar to asthma; the others had only episodes of apnoea. Wistar and Long–Evans rats resembled Sprague–Dawley rats; Fischer 344 rats had apnoea only, even though they produced IgE antibodies. The type of response did not correlate with serum IgE levels. The respiratory responses were reduced by dexamethasone, disodium cromoglycate, epinephrine and theophylline. Rats that respond with dyspnoea may provide a useful experimental model of allergic asthma.

INTRODUCTION

It is generally accepted that human allergic asthma is caused by an immediate hypersensitivity reaction which is mediated by IgE-type reaginic antibodies. Appropriate animal models of allergic asthma (for review see Patterson & Kelly, 1974) are necessary to allow investigations of the mechanisms involved and to assist in the assessment of potential new drugs. Many studies of allergic asthma in dogs (Krell, Chakrin & Wardell, 1975; Kepron, James, Kirk, Sehon & Tse, 1977; Faith, Hessler & Small, 1977) and monkeys (Patterson, Harris, Suszko & Roberts, 1976) have been reported but these species have disadvantages which preclude their routine use. Small animal models of allergic asthma would have distinct advantages. Guinea-pigs undergo intense anaphylactic bronchoconstriction, but their response differs from human allergic asthma both in the immunoglobulins involved and in the inconsistent effect of disodium cromoglycate (Carney, 1976).

Following appropriate sensitization, rats produce IgE antibodies (Stechschulte, Orange & Austen, 1970) which are similar to human reaginic antibody. Anaphylactic bronchoconstriction mediated by IgE has been described in rats infected with *Nippostrongylus brasiliensis* (Church, Collier & James, 1972), actively sensitized against ovalbumin (Stotland & Share, 1974a) and passively sensitized with antibodies to conalbumin (Farmer, Richards, Sheard & Woods, 1975). Although pulmonary reactivity to antigen challenge was measured, the rats were either pithed or anaesthetized and the antigen was administered intravenously.

This investigation was initiated to determine the effect of aerosolized antigen on conscious sensitized rats. A procedure was used which resulted in a respira-

* Presented in part at the 21st Annual Meeting of the Canadian Federation of Biological Societies, London, Ontario, June 1978.

Correspondence: Dr G. Holme, Pharmacology Department, Merck Frosst Laboratories, P.O. Box 1005, Pointe Claire-Dorval, Quebec H9R 4P8, Canada.

0019-2805/79/1000-0385$02.00

tory response which resembled asthma and which was sensitive to drugs. Another study was recently described (Carswell & Oliver, 1978a) in which aerosolized antigen produced a respiratory response, but it was not as pronounced as that reported here.

MATERIALS AND METHODS

Materials

Egg albumin (EA), grade V, salt free, crystallized and lyophilized and bovine γ-globulins (BGG), Cohn fraction II, were obtained from Sigma Chemical Co., St Louis, U.S.A. Aluminium hydroxide was supplied by the Reheis Chemical Company, Chicago, U.S.A. The following pharmacological agents were used: dexamethasone 21-phosphate disodium, Merck and Co. Inc., Rahway, U.S.A.; disodium cromoglycate, Fisons Ltd, Loughborough, U.K.; epinephrine bitartrate, Sigma Chemical Company, St Louis, U.S.A.; theophylline, K and K Laboratories Inc., Hollywood, U.S.A.

Bordetella pertussis vaccine, containing 15×10^9 killed bacilli/ml, was supplied by Connaught Medical Research Laboratories, Toronto, Canada. Rats infected with *N. brasiliensis* were obtained from Dr K. Bloch, Harvard University, Boston, U.S.A. The parasite was maintained in rats by infecting with larvae and collecting the eggs. Suspensions of larvae were prepared as described by Keeling (1960).

Animals

Sprague–Dawley, Long–Evans and Wistar rats, female 200–250 g, were supplied by Biobreeding Laboratories, Ottawa, Canada. Fischer 344 rats, female 150–180 g, were from Canadian Breeding Farms, St Constant, Quebec, Canada. Hartley guinea-pigs, male 300–400 g, were supplied by Connaught Medical Research Laboratories, Toronto, Canada.

Immunization procedures

Rats were routinely immunized by injecting (s.c.) 1 ml of a suspension containing 1 mg EA (or BGG) and 200 mg aluminum hydroxide and injecting (i.p.) simultaneously 1 ml of *B. pertussis* vaccine. Rats were usually challenged 14–18 days later. When *N. brasiliensis* was included in the immunizing regimen, 4×10^3 larvae in 1 ml of saline were injected (s.c.) on day 10 and rats were challenged 10–14 days later. Immunization by aerosol was carried out by injecting (i.p.) 1 ml of *B. pertussis* vaccine and then exposing the rats for 5 min to an aerosol of 1% EA in saline. Passive sensitization of rats was accomplished by injecting (i.v.) 4 ml of rat serum with a PCA titre of 256. The serum, from rats immunized with EA, aluminum hydroxide and *B. pertussis*, was injected in 1 ml portions over a period of 6–8 h. Samples of the same serum were heated at 56° for 3 h before injection. Rats were challenged 72 h after passive sensitization. Guinea pigs were sensitized to EA by a method based on that of Margni & Hajos (1973). Each animal received 1 ml of 0·1% EA in saline (i.m.) and 1·0 ml *B. pertussis* vaccine (i.p.). The injections were repeated on day 7 and the animals were used on day 12.

Serum IgE titres

Serum IgE levels were determined by passive cutaneous anaphylaxis (PCA). Serial two-fold dilutions of the test serum were made in saline. An intracutaneous injection of 0·05 ml of each dilution was then made into the dorsal region of the recipient rat. Duplicate determinations were carried out using two recipients. After 72 h the rats were challenged by injecting (i.v.) 1 ml of a solution containing 5 mg EA and 0·5% Evans's blue in saline. The diameters of the blue wheals on the skin were measured 30 min after challenge. The PCA titre was recorded as the reciprocal of the highest dilution that produced a wheal at least 5 mm in diameter. Heating the serum at 56° for 3 h markedly reduced the PCA titre.

Aerosol challenge

The challenge and subsequent recording were carried out in a clear plastic box with internal dimensions $10 \times 6 \times 4$ inches. The top of the box was removable to allow access; in use, it was held firmly in place by four clamps. An airtight seal was maintained by a soft rubber gasket around the top edge of the chamber. A ¼ in stainless steel wire mesh platform was mounted 1 in above the floor of the box. Through the centre of each end of the chamber a De Vilbiss nebulizer (No. 40) was inserted and an airtight seal was maintained by a rubber collar. Each end of the box also had an outlet. Into one end of the box a Fleisch No. 0000 peumotachograph was connected through a short length (1 in) of polypropylene tubing. The connector, tubing and pneumotachograph had similar (6 mm) internal diameters. The pneumotachograph was coupled to a Grass volumetric pressure transducer which was connected to a Beckman Type R Dynograph through appropriate couplers. While aerosolizing the antigen, the outlets were open and the pneumotachograph was isolated from the chamber by clamping the connecting

tubing. The outlets were closed and the connection between the pneumotachograph and the chamber was open during the recording of the respiratory patterns. Thus, during the recording phase all air exchanges between the chamber and its surroundings were through the pneumotachograph.

For exposure to the aerosol and subsequent recording of the respiratory patterns, the rats (or guinea-pigs) were placed in the box and the top was tightly sealed. Throughout the experiment animals were conscious, unrestricted and free to move around in the chamber. Normally for challenge, 2 ml of a 3% solution of antigen in saline was placed into each nebulizer. The aerosol was generated with air from a small Potter diaphragm pump operating at 10 psi and a flow of 8 l/min. Fresh antigen solution was used for challenging each animal and saline was used for controls. Rats were exposed to the aerosol for exactly 1 min and guinea-pigs for exactly 30 s before the respiratory patterns were recorded for 15 min. The respiratory pattern of each animal was monitored (15 s) before challenge.

When the respiratory pattern of an animal was being recorded, two air-exchange processes occurred simultaneously. One was the inspiration and expiration of air by the rat from and into the chamber space. The other process was similar to that occurring in a plethysmograph and involved the expulsion and uptake of air between the chamber and its surroundings due to the animals respiratory movements. The summation of the two air exchanges due to these two processes caused the resultant air flow between the chamber and its surroundings. The pneumotachograph quantified the resultant flow which was recorded to produce the respiratory patterns. Thus, the respiratory patterns do not solely record either the rat's tidal flow or, as with a body plethysmograph, the air exchange between chamber and surroundings due to respiratory movement.

Drug evaluations

Epinephrine (0·03 mg/kg) or disodium cromoglycate (30 mg/kg) were administered (i.v.) to rats in saline 1 min prior to challenge. Theophylline (100 mg/kg) was administered (p.o.) as a fine suspension in 1% methocel 30 min prior to challenge. Dexamethasone (10 mg/kg) in saline was administered (p.o.) twice at 18–20 h and again 1 h before challenge. For controls, similar groups ($n=10$) of rats were treated with the same volume of solvent and then challenged. The response to challenge was quantitated with reference to two parameters. First, the duration (min) of continuous dyspnoea (Fig. 1C) was measured from the recording chart corresponding to each rat and a group mean was calculated. Second, the frequency (number/min) of episodes of apnoea (Fig. 1A) were counted from the recording chart. In computing frequency, only the time (min) when the rats were actually experiencing episodes of apnoea was used; the time was not included when continuous dyspnoea or no symptoms occurred. The Student's two-tailed t test was used to determine the significance of differences between treated and control groups.

RESULTS

Prior to challenge, the rat's respiratory pattern (Fig. 1) was dependent on whether it was quiescent or exploring. Following exposure to antigen aerosols, distinct changes in the patterns occurred in about 90% of the

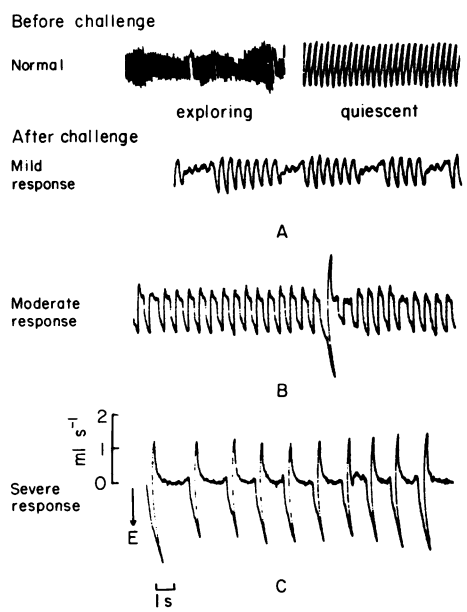

Figure 1. Photograph of segments of the experimental record of rat respiratory patterns before and after challenge with an aerosol of ovalbumin. The direction of flow during the expiratory phase (E) and a 2 ml/s calibration are shown. The recording speed is indicated by a 1 s interval. N.B. The pattern represents the resultant air flow into or from the respiratory chamber due to respiration. It indicates qualitative differences in the respiration and does not record tidal flow, so pulmonary parameters cannot be quantified directly. A more detailed explanation is given in the methods section.

rats. The least change in the pattern (mild response), which was shown by all rats that responded, resulted in periods of apnoea (Fig. 1A) and a respiratory rate similar to that of a quiescent rat. The rats showed no evident discomfort even during periods when episodes of apnoea were being recorded. After the initial phase, a number of rats (a proportion variable from 20 to 70%) developed a greater change in the respiratory pattern (moderate response). This pattern (Fig. 1B) showed a slight delay before the expiratory phase, an occasional cycle of increased flow and some episodes (not shown) of apnoea similar to Fig. 1A. During this phase, pronounced movements of the thoracic and abdominal muscles were observed.

With a few exceptions, rats whose response progressed beyond the initial phase then developed even greater changes in the respiratory pattern (severe response). There was a marked decrease in respiratory rate and each cycle was characterized by increased flow and a pronounced delay before the onset of the expiratory phase (Fig. 1C). Rats experiencing this response had continuous dyspnoea and evident distress. They assumed a pronounced stance, with fore and hind limb extension, there was exaggerated movement of the thoracic and abdominal muscles and obvious gasping. Bluish colouration of the tongue, extremities and mucous membranes indicated cyanosis and some rats became prostrate. The respiratory patterns and other symptoms observed during the severe response resembled those seen when sensitized guinea-pigs were challenged in a similar manner. The guinea-pig respiratory pattern (not shown) was similar to Fig. 1C but was characterized by a more prolonged delay in the onset of the expiratory phase. The delay became progressively longer until the guinea-pig died after 2–3 min.

The mild response occurred within 1 min after exposure to antigen; the moderate or severe responses started 2–5 min after challenge. Each response continued for several minutes before the rats recovered. As rats recovered from the severe response, their breathing patterns first resembled Fig. 1B then Fig. 1A and eventually, after several minutes, they returned to normal.

No respiratory response was elicited from sensitized rats by challenge with saline. Rats sensitized to EA and challenged with BGG showed no significant changes in their breathing patterns. The same rats challenged 24 h later with EA had respiratory responses; five rats had a mild response and ten had a severe response. Similar results were obtained when rats sensitized to BGG were challenged first with EA and then with BGG. Sensitized rats were routinely exposed to aerosolized antigen between days 14 and 18 post-sensitization, but their susceptibility to challenge was of longer duration. At various times between day 8 and day 63 post-sensitization, rats responded to challenge; each group comprised mild, moderate and severe responders. Passive sensitization of rats by i.v. injection of serum containing EA-specific IgE produced a respiratory response in the recipients following aerosolized antigen challenge. The response was absent if the serum was heated before injection (Table 1).

Different EA solutions, from 0·3 to 10%, were used in aerosols to challenge groups ($n=10$) of rats but none produced a homogeneous response in which all rats had continuous dyspnoea. Groups of rats ($n=10$) were sensitized by four procedures which were expected to produce different IgE levels. The results (Table 2) show that the procedure which produced the highest serum IgE titre did not increase the proportion of severe responders. These data were used in Fig. 2 to plot duration of dyspnoea against IgE level. Each point on the abscissa refers to a rat which had either a mild or moderate response. Four different strains of rats were examined but none comprised a homogeneous population in which all the rats had continuous dyspnoea following challenge. Wistar and Long–Evans rats resembled Sprague–Dawley rats in that only a proportion of the rats had a severe response to antigen challenge (Table 3). Fischer 344 rats did not have continuous dyspnoea even though their sera contained IgE antibodies to EA.

To determine the effect of drugs, the respiratory

Table 1. The effect of passive sensitization on the respiratory response to aerosolized antigen challenge

Serum	Rat No.	Response
Not heated	1	Servere: duration 3·3 min
	2	Mild
	3	Severe: duration 3·6 min
	4	Mild
Heated 56°, 3 h	5	None
	6	Slight*
	7	Slight*
	8	None

Serum: 4 ml with a PCA titre of 256.

* The respiratory pattern was different to normal but not as pronounced as A (Fig. 1).

Table 2. The effect of method of sensitization on PCA titre and on response to aerosolized antigen challenge

Method of sensitization	Range of PCA titre	Severe responders* %
1% EA aerosol 5 min + *B. pertussis* i.p.	2–4	38
1 mg EA + 200 mg Al $(OH)_3$ s.c.	4–32	50
1 mg EA + 200 mg Al $(OH)_3$ s.c. + *B. pertussis* i.p.	16–256	40
1 mg EA + 200 mg Al $(OH)_3$ s.c. + *B. pertussis* i.p. + *N. brasiliensis* s.c.	64–512	24

Challenge: 3% EA aerosol for 1 min. Groups of ten rats were tested.
B. pertussis $1{\cdot}5 \times 10^9$ cells. *N. brasiliensis* 4×10^3 larvae.
* Severe responders (%) is the proportion of animals in a test group ($n = 10$) which had respiratory patterns like Fig. 1C following antigen challenge.

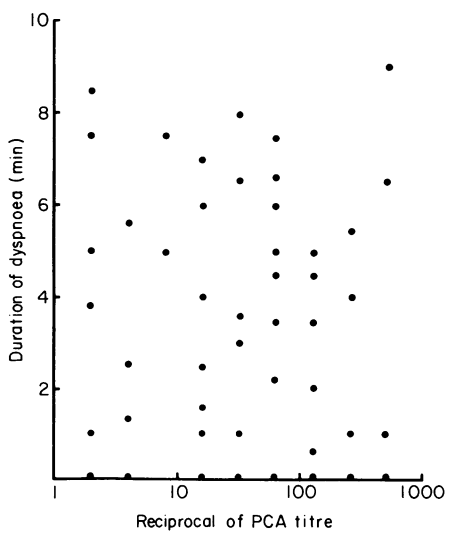

Figure 2. Comparison of duration of dyspnoea and PCA titre following challenge of rats with antigen aerosol. The points on the abscissa refer to rats which did not exhibit dyspnoea following challenge.

response was quantified in terms of both the mild and severe responses by determining the frequency of apnoea and the duration of continuous dyspnoea, respectively. The results are shown in Table 4. Dexamethasone (10 mg/kg) and epinephrine (0·03 mg/kg)

Table 3. The proportion of severe responders in different strains of rats

Strain	PCA titre	Severe responders* (%)	No of groups tested
Sprague–Dawley	2–512	20–70	20
Wistar	—	20, 20	2
Long–Evans	—	30, 50	2
Fischer 344	4–64	0	5

Challenge: 3% EA aerosol for 1 min. Groups contained ten rats.
PCA titre is the reciprocal of highest dilution of serum which results in a 5 mm diameter wheal on the recipient.
* See note to Table 2.

completely prevented continuous dyspnoea while disodium cromoglycate (30 mg/kg) and theophylline (100 mg/kg) caused a 93% and 80% inhibition, respectively. The drugs reduced the frequency of apnoea to a lesser extent, ranging from 33% with disodium cromoglycate to 64% with dexamethasone.

DISCUSSION

Experimental asthma in rats has generally been investigated using either anaesthetized or pithed animals which were then challenged by intravenous administration of the antigen. The present investigations were initiated to determine the effect of challenging conscious rats with an aerosol of the sensitizing antigen. The recordings of the subsequent respiratory patterns (Fig. 1) show that significant changes occurred following exposure to aerosolized antigen. Another investigation of the response of conscious rats (Carswell & Oliver, 1978a) used a different experimental procedure in which the rats were restrained in a constant volume body plethysmograph. Although changes in the respiratory patterns, termed 'expiratory notching', were observed, they were not as pronounced as those shown in Fig. 1. The respiratory response occurred only after challenge with the sensitizing antigen indicating that it required a specific antibody/antigen interaction; aerosols of either saline or a protein different to the sensitizing antigen elicited no response. The rats responded to challenge even after 63 days post-sensitization, suggesting that IgE antibodies were responsible because they are known to be persistent at tissue sites (Binaghi & Benacerraf, 1964). Furthermore, non-immunized

Table 4. Effect of drugs on the respiratory response of rats to aerosolized antigen challenge

Treatment	Dose (mg/kg)	Mild response† frequency of apnoea (episodes/min)	Severe response‡ duration dyspnoea (min)
Control	—	7·2 ± 2·1	4·2 ± 2·9
Disodium cromoglycate	30	4·9 ± 1·6	0·3 ± 0·9
Control	—	2·2 ± 0·5	4·1 ± 2·3
Theophylline	100	1·7 ± 0·8*	0·8 ± 1·3
Control	—	2·8 ± 1·5	4·9 ± 4·1
Dexamethasone	10	1·1 ± 0·7	0
Control	—	3·8 ± 1·0	2·4 ± 2·6
Epinephrine	0·03	2·6 ± 1·0	0

Disodium cromoglycate and epinephrine were administered i.v. and theophylline and dexamethasone were administered p.o.
Difference between treatment mean and control mean were significant ($P < 0·05$) except where indicated (*).
† The mild response and episodes of apnoea are illustrated in Fig. 1A.
‡ The severe response and dyspnoea are illustrated in Fig. 1C.

rats were passively sensitized to respond to challenge by injecting serum from sensitized rats. The sensitizing activity of the serum was abolished by prior heating thus providing further support for the conclusion that IgE antibodies mediated the response.

The response by rats to aerosolized antigen was variable. Each group contained rats which showed each type of response and also rats which did not respond. The majority, however, comprised rats having either mild or severe responses. Heterogeneity in response to aerosolized antigen was not noted in other investigations with conscious rats (Carswell & Oliver, 1978a). The response which included continuous dyspnoea (Fig. 1C) was the most severe, particularly in view of the associated symptoms such as cyanosis and prostration. The acute respiratory distress that guinea-pigs experience on exposure to aerosolized antigen is an accepted experimental model for respiratory anaphylaxis (Patterson & Kelly, 1974). The respiratory profile of rats during the severe response was almost identical to the breathing pattern of guinea-pigs following challenge. This suggests that rats responding with continuous dyspnoea could be used as an alternative model of allergic asthma. Determinations of changes in pulmonary mechanics parameters would demonstrate how the response compares with human asthma.

The basic experimental procedure was changed in attempts to obtain a homogeneous response in which all rats responded with continuous dyspnoea. The heterogeneity is unlikely to be due to individual rats receiving different amounts of EA because challenge with solutions containing concentrations of antigen (0·3–10%) yielded examples of each type of response. The experiments in which different sensitizing procedures were used allowed the response to be evaluated in rats whose serum had PCA titres ranging from 2 to 512. Note that a single exposure to aerosolized EA was sufficient to elicit IgE production, confirming the findings of Van Hout & Johnson (1972). The sensitizing procedure which produced the highest IgE titres yielded the lowest proportion of severe responders, suggesting that serum IgE levels do not determine the type of response. The lack of correlation is evident in Fig. 2 where duration of dyspnoea is plotted against PCA titre. Continuous dyspnoea of similar duration was observed in rats whose serum IgE titres ranged from 2 to 512. Other rats with similar IgE titres had mild or moderate responses only. The lack of correlation between serum IgE levels and the respiratory response in rats has been shown previously (Carswell & Oliver, 1978a; Church, 1975). In dogs, bronchoprovocation elicited a response only when serum IgE levels were above a threshold level (Kepron *et al.*, 1977).

The heterogeneous respiratory response was also characteristic of Wistar and Long–Evans rats (Table 3). Two groups from each strain comprised rats which

had mild severe or no responses. The most interesting finding was that five groups (total fifty rats) of Fischer 344 rats had only mild responses to antigen challenge. The absence of dyspnoea was not due to their inability to make IgE antibodies to EA because they had serum titres that had been associated with severe responses in Sprague–Dawley rats (Table 2). Thus, Fischer 344 rats may represent a population which have a predictably mild response to challenge and they could be used to characterize that response more fully. The respiratory patterns (Fig. 1A) of mild responders, though distinct from those of rats prior to challenge, may represent a minimal change in pulmonary parameters. Determinations of pulmonary mechanics parameters with Fischer 344 rats would show the magnitude of the response. If it is small, then it would be reasonable to consider Sprague–Dawley rats being comprised of responders (i.e. those with post-challenge respiratory patterns as Fig. 1C) and non-responders (i.e. those with post-challenge patterns as Fig. 1A). Dogs (Krell *et al.*, 1975) and rhesus monkeys (Patterson & Talbot, 1972) have also been segregated into responders and non-responders depending on the magnitude of antigen-induced changes in pulmonary parameters.

The physiological and immunological aspects of animal models of asthma have been studied extensively but few pharmacological studies have been reported (Krell *et al.*, 1975), probably due to the variability in response between individual animals and between successive challenges of the same animal. It was expected that precise dose-related effects of drugs would be difficult to obtain in view of the heterogeneity in response of the rats. Nevertheless, a preliminary characterization was obtained by administering drugs at a dose that was expected to have a substantial effect. Catecholamines and theophylline are important drugs in the clinical management of asthma due to their spasmolytic effects and their activity in inhibiting mast cell degranulation (Taylor, Francis, Sheldon & Roitt, 1974). Their effect in inhibiting the respiratory impairment, particularly the continuous dyspnoea, indicates it was due to constriction of airways smooth muscle by released mediators. The dose of theophylline used is comparable to the dose used to reverse allergic bronchoconstriction in cats (Barch & Talbot, 1976); epinephrine has been used at similar doses to reverse bronchoconstriction in dogs (Gold, Kessler, Yu & Frick, 1972). Disodium cromoglycate inhibits mediator release by preventing mast cell degranulation (Church, 1978). Its effect on the respiratory impairment, together with the involvement of IgE, is consistent with the response being due to contractile mediators released by the interaction of cell-bound IgE and antigen. Disodium cromoglycate also caused a significant reduction of the anaphylactic bronchoconstriction in anaesthetized (Stotland & Share, 1974b) or pithed (Church *et al.*, 1972) rats following i.v. antigen challenge. The intratracheal administration of disodium cromoglycate (6 mg) also reduced the 'expiratory notching' (Carswell & Oliver, 1978b) that followed challenge of conscious rats with aerosolized antigen. Corticosteroids are used clinically for the management of asthma that is intractable to other drugs. The inhibition of the respiratory response by dexamethasone is in agreement with the findings of Church *et al.* (1972), who showed a dose-dependent inhibition of anaphylactic bronchoconstriction in the rat. Other studies (Stotland & Share, 1974b) indicated that a more prolonged treatment with dexamethasone was required for inhibition of bronchial anaphylaxis in rats.

The drugs were more effective in minimizing the duration of dyspnoea than in reducing the frequency of apnoea. The probable explanation is that limiting drug concentrations attenuated an otherwise severe response (dyspnoea) so that it was manifested as a mild response (apnoea). In those rats which otherwise would have had a mild response only, the frequency of the symptoms was reduced but not eliminated. The drug results thus support the observations that antigen challenge elicits two distinct responses with the one that includes continuous dyspnoea being the more severe.

There would be distinct advantages to having a homogeneous population in which all rats had continuous dyspnoea in response to challenge. They could be used to assess the utility of new drugs, determine changes in pulmonary mechanics in comparison with Fischer 344 rats and investigate mechanisms involved in asthma. Rhesus monkeys are also variable in their response to aerosolized ascaris antigen but the responders are hyper-reactive to inhaled carbocholine and this property has been used to distinguish them from non-responders (Patterson *et al.*, 1976). A linkage between the severe response and another characteristic which could be used as a marker has not been investigated but is of interest for future studies. The breeding characteristics and life span of rats, however, make selective breeding an alternative approach to obtaining a homogeneous population. An inbred line of rats which do not have an anaphylactic response to dextran has been developed by selective breeding from

Wistar rats (Harris, Kalmus & West, 1963). Non-responsiveness was regulated by an autosomal, recessive gene which could be outbred into other stock. Such an approach to obtaining a homogeneous population of rats that respond to antigen aerosols with dyspnoea may also allow the genetic characteristics of asthma to be investigated.

REFERENCES

Barch G.K. & Talbott M.W. (1976) Allergic bronchoconstriction and its drug-induced reversal in anaesthetized, ovalbumin-sensitized cats. *Res. Comm. Chem. Path. Pharmacol.* **13,** 623.

Binaghi R.A. & Benacerraf B. (1964) The production of anaphylactic antibody in the rat. *Immunology,* **92,** 920.

Carney I.F. (1976) IgE-mediated anaphylactic bronchoconstriction in the guinea pig and the effect of diosdium cromoglycate. *Int. Archs Allergy appl. Immun.* **50,** 322.

Carswell F. & Oliver J. (1978a) The respiratory response in sensitized rats to aerosol challenge. *Immunology,* **34,** 465.

Carswell F. & Oliver J. (1978b) Site of respiratory reaction in allergic rats challenged via the airways. *Int. Archs Allergy appl. Immun.* **57,** 358.

Church M.K. (1975) Correlation of anaphylactic bronchoconstriction with circulating reaginic antibody level and active cutaneous anaphylaxis in the rat. *Immunology,* **29,** 527.

Church M.K. (1978) Cromoglycate-like anti-allergic drugs: a review. *Med. Act./Drugs of Today,* **14,** 281.

Church M.K., Collier H.O.J. & James G.W.L. (1972) The inhibition by dexamethasone and disodium cromoglycate of anaphylactic bronchoconstriction in the rat. *Br. J. Pharmacol.* **46,** 56.

Faith R.E., Hessler J.R. & Small P.A. (1977) Respiratory allergy in the dog: induction by the respiratory route and the effect of passive antibody. *Int. Archs Allergy appl. Immun.* **53,** 530.

Farmer J.B., Richards I.M., Sheard P. & Woods A.M. (1975) Mediators of passive lung anaphylaxis in the rat. *Br. J. Pharmacol.* **55,** 57.

Gold W.M., Kessler G.F., Yu D.Y.C. & Frick O.L. (1972) Pulmonary physiologic abnormalities in experimental asthma in dogs. *J. appl. Physiol.* **33,** 496.

Harris J.M., Kalmus H. & West G.B. (1963) Genetical control of the anaphylactoid reaction in rats. *Genet. Res.* **4,** 346.

Keeling J.E.D.(1960) The effects of ulta-violet radiation on *Nippostrongylus muris.* I. Irradiation of infective larvae: lethal and sublethal effects. *Ann. trop. Med. Parsitol.* **54,** 182.

Kepron W., James J.M., Kirk B., Sehon A.H. & Tse K.S. (1977) A canine model for reaginic hypersensitivity and allergic bronchoconstriction. *J. Allergy clin. Immunol.* **59,** 64.

Krell R.D., Chakrin L.W. & Wardell J.R. (1975) *In vivo* canine and rhesus monkey models of allergic asthma. In: *Immunopharmacology* (Ed. by M. E. Rosenthale and H. C. Mansmann), p. 125. Spectrum Publications Inc., New York.

Margni, R.A. & Hajos S.E. (1973) Guinea-pig reaginic antibody. II. Physicochemical and biological properties. *Immunology* **25,** 333.

Patterson R., Harris K.F., Suszko I.M. & Roberts M. (1976) Reagin-mediated asthma in rhesus monkeys and relation to bronchial cell histamine release and airway ractivity to carbocholine. *J. clin. Invest.* **57,** 586.

Patterson R. & Talbot C. (1972) A comparison of immediate-type respiratory reactions to immunologic and pharmacologic agents in rhesus monkeys. *J. Allergy clin. Immunol.* **49,** 292.

Patterson R. & Kelly J.R. (1974) Animal models of the asthmatic state. *Ann. Rev. Med.* **25,** 53.

Stechschulte D.J., Orange R.P. & Austen K.F. (1970) Immunochemical and biologic properties of rat IgE. *J. Immunol.* **105,** 1082.

Stotland L.M. & Share N.N. (1974a) Active bronchial anaphylaxis in the rat. *Can. J. Physiol. Pharmacol.* **52,** 1114.

Stotland L.M. & Share N.N. (1974b) Pharmacological studies on active bronchial anaphylaxis in the rat. *Can. J. Physiol. Pharmacol.* **52,** 1119.

Taylor W.A., Francis D.H., Sheldon D. & Roitt I.M. (1974) Anti-allergic actions of disodium cromoglycate and other drugs known to inhibit cyclic 3′, 5′-nucleotide phosphodiesterase. *Int. Archs Allergy,* **47,** 175.

Van Hout C.A. & Johnson H.G. (1972) synthesis of rat IgE by aerosol immunization. *J. Immunol.* **108,** 834.