Learned Asthma in the Guinea Pig

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THERE ARE FEW EXPERIMENTAL reports substantiating the hypothesis that bronchial asthma can be a learned phenomenon. It has been suggested by French and Alexander[5] and others[18] that one of the psychological factors involved in bronchial asthma is analogous to the conditioned reflex. Stimuli that have been regularly associated with the presence of an allergic substance may precipitate an attack in a susceptible individual.

Conducting allergic studies with humans, Herxheimer[7] observed that merely placing many of the asthmatic subjects in a situation in which on repeated occasions they had been exposed previously to allergens, they developed asthma without the exposure. Dekker, Pelser, and Groen[3] report conditioned asthmatic attacks in a laboratory setting. They followed a procedure similar to Herxheimer's, using a known allergen paired with neutral solvents, and found in two subjects after repeated trials that the inhalation of pure oxygen caused attacks of asthma as demonstrated by clinical signs and vital capacity. The mouthpiece alone was eventually sufficient to cause an attack. These attacks could not be distinguished from those that appeared after the allergen. Dekker and Groen[2] also report psychogenic attacks of asthma using stressful stimuli that have in the past precipitated attacks in their subjects. They concluded that this evidence seemed in line with a conditioning mechanism although it did not follow a classical example.

Liddell[9] has surveyed the literature on conditioning of respiration in animals and its psychosomatic implications in humans. When experimental neuroses are produced in animals by conditioned reflex mechanisms there appear changes in the rate, rhythm, and pattern of respiration. Gantt[6] produced in an experimentally neurotic dog "loud, raucous breathing with quick inspiration and labored expiration accompanied by a loud wheezing." This respiratory pattern appeared when the dog was brought from the paddock, became more pronounced as the experimental situation was approached, and disappeared in reverse order. This reaction persisted for many years. Examination of this dog showed that there was no true bronchiolar constriction.

Liddell reported that respiratory dysfunction is an invariable manifestation of chronic experimental neuroses in animals and resembles the labored breathing of bronchial asthma in some instances.

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Another approach to the experimental study of asthma is the use of classical conditioning techniques. Such investigation is feasible since Ratner[14] and others[8] have demonstrated that a finely sprayed antigen produces in sensitized guinea pigs asthmatic attacks which resemble human bronchial asthma. In 1927 Ratner introduced a method for sensitizing and inducing anaphylaxsis in guinea pigs solely through the inhalation of dry antigenic dust. The symptom complex produced he called "experimental asthma" because it was comparable to the asthmatic syndrome in the human. Kallos and Pagel[8] concluded from their work that finely sprayed antigen produces in sensitized guinea pigs attacks that clinically, roentgenologically, pharmacologically, and immunologically resemble human bronchial asthma. Ratner[15] has recently concluded that the inhalation of aerosolized liquids and dust results in reactions that are to all intents and purposes similar to human bronchial asthma.

Noelpp and Noelpp-Eschenhagen[11, 12] have reported conditioning experiments with guinea pig asthma in which an allergic attack was paired with auditory stimuli. After 5 conditioning trials at irregular intervals, 1 of 8 animals had an asthmatic type of respiration in response to the conditioned stimulus. An additional 5 conditioning trials resulted in conditioned asthmatic responses in 3 more of the animals.

This study deals with the further investigation of experimental and learned asthma in the guinea pig.

Method

The experiment was divided into two parts. Part I was concerned with the development of experimental asthma in the guinea pig; Part II was concerned with obtaining information about learned asthmatic responses.

Part I

Thirty young male guinea pigs weighing 200-300 Gm., were injected intraperitoneally 3 times with 0.25 ml. of fresh, undiluted egg white in a 1-week period. Following the series of injections 2 weeks were allowed to elapse for sensitization to occur. A special chamber, 10″ × 16″ × 16″ was constructed with a one-way-vision window to permit observation of the animal's respiratory patterns. The chamber had an aperture for a #15 De Vilbiss nebulizer attached to a constant-pressure and -flow air pump. The nebulizer served to introduce a fine mist of a dilute solution of egg white. The animals were placed individually in the chamber on consecutive days. After 1 minute of observation the egg-white spray was introduced. Duration of the period in the chamber was 15 minutes during the early trials but often of shorter duration in the later trials because the severity of the respiratory distress necessitated removal of the animal. The box was thoroughly cleansed following each trial to remove residual egg white. Four other sensitized animals given only a saline spray served as a control group.

Two judges familiar with the procedure timed these attacks. An attack was marked by use of accessory muscles, gasping, coughing, and pronounced respiratory distress.

Part II

The 6 animals who had attacks on 10 or more consecutive trials were placed daily in the chamber in the absence of the egg-white spray. Independent observers noted the respiratory patterns and judged the presence or absence of an asthmatic attack, as in Part I. The animals were tested daily until no attacks occurred in 15 minutes on 5 consecutive daily trials, at which time the response was considered extinguished.

Results

Part I

The guinea pigs previously made sensitive to egg white developed respiratory distress when exposed to the nebulized egg-white spray. In producing the respiratory attack it was found that the components developed in a fairly uniform fashion. The

sequence of respiratory signs was as follows: rapid breathing, restlessness, chewing, ruffling of the fur, then labored breathing with the use of accessory muscles of respiration, prolonged expiration, gasping, coughing, dilation of alae nasi, and finally cyanosis and convulsion. The severity in individual asthmatic attacks varied and all of the animals did not have attacks on each trial. All of the animals demonstrated sensitivity to the egg-white spray in the initial exposure, but after the second trial less than half continued with respiratory distress (Fig. 1).

As the experiments progressed there was an increasing delay between the exposure to antigen and the onset of the asthmatic attack. The difference between the time of the first and the third attack was highly significant ($P < 0.001$). Only 20 per cent of these animals had daily attacks. Mean latency time of the onset of their attacks was much shorter than that of the entire group (Fig. 2). In contrast, the control animals exposed only to the saline spray did not develop any of the signs of respiratory distress.

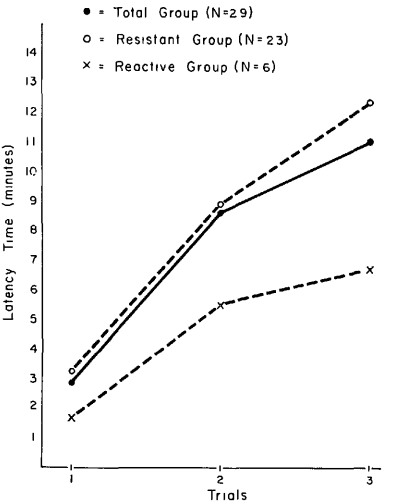

Fig. 2. The mean time between exposure to antigen and onset of attacks for the first 3 trials. The reactive group was composed of the 20 per cent of the animals who had 13 daily attacks. The resistant group composed the remainder.

Part II

During the extinction phase all of this 20 per cent (6 animals) had asthmatic attacks without the presence of egg-white spray. Four animals continued to have attacks through 9 trials. The attacks were milder and did not progress to convulsive trembling or seizures. Figure 3 showing

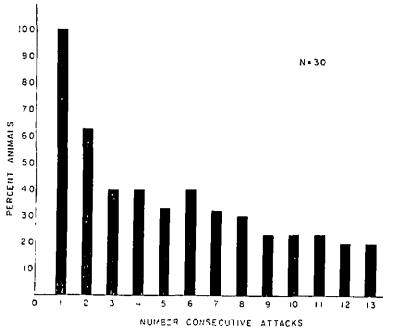

Fig. 1. The percentage of animals and number of consecutive attacks.

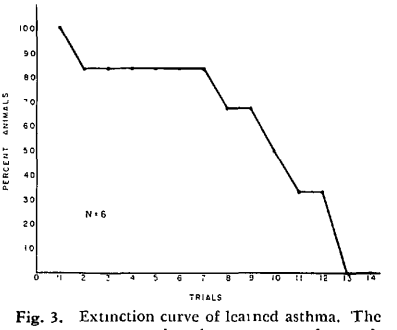

Fig. 3. Extinction curve of learned asthma. The responses are expressed as the percentage of animals who had learned asthmatic attacks on consecutive daily trials.

the extinction curve for the animals follows a typical extinction pattern in response to a learned stimulus. Extinction was apparent in all after 13 trials. This we define as learned asthma.

Discussion

The present study suggests that there may be varying degrees of susceptibility to asthma in different animals, a fact which is important in selecting the most sensitive populations for further psychological experiments. Only 20 per cent of the initial group of animals had consistent daily attacks. The range in the time of onset of attacks also varied, with the most reactive animals having shortest latency periods.

The processes involved in the variability of latency and number of repeated asthmatic attacks is not clear and may be related to immunological changes of desensitization and antianaphylaxis. In another experiment Ottenberg, Stein, and Lewis[13] found that in sensitized guinea pigs single or repeated attacks often lead to a latency time exceeding 15 minutes. Allowing these animals 2-5 days of nonexposure to egg white results in a reduction of the latency period and a recovery of their reactivity.

In a third study 10 animals were exposed to the same procedure with several modifications. A much smaller chamber (15 per cent of the original volume) was utilized to reduce the activity of the animal and to increase the concentration of egg white in the atmosphere. From Fig. 4 it can be seen that all 10 of the guinea pigs had 10 consecutive attacks like the 20 per cent of the group in the larger chamber. The increased reactivity may be due to the higher concentration of antigen; or it could be due to higher susceptibility of the subjects.

It has been suggested that guinea pigs vary in their capacity to become sensitized. Ratner[16, 17] has found that 7 per cent of guinea pigs are resistant to the development of experimental asthma and suggests that this may be an innate factor. Zinsser[19] in an earlier study confirmed the finding of resistant strains. Resistance does not appear to be a major factor in the present study, since all of the animals had attacks on the initial trial. Feinberg[4] has demonstrated that the time of onset of asthmatic attacks in guinea pigs is influenced by prior treatment with ACTH, cortisone, and stress.

The difference in the reaction of guinea

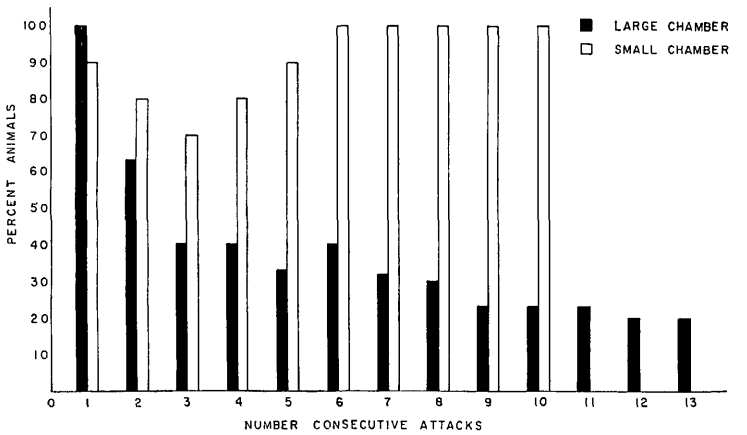

Fig. 4. Influence of chamber size on number of consecutive attacks.

pigs to the injection and the inhalation of egg-white spray may be related to immunological, genetic, hormonal, local, or psychological variables. Our findings suggest that those that react most quickly to the allergen can be identified very early in the experiment. The findings of differences in the number of repeated attacks and changing latencies in animals are consistent with Mirsky's[10] concept that inherent biological factors play a large role in the susceptibility to disordered function.

This methodology for the production of experimental asthma in a susceptible group afforded an opportunity to investigate learned asthma in the absence of an allergen. Results of the present study indicate that asthma can be learned. Of the 6 susceptible animals, 4 had nine daily attacks in response to the chamber without allergen. The question might be raised that the extinction curve represents merely the physiological process of removal of antigen from the animal's system. The concentration of egg-white mist was heavy, and some observers have suggested that allergen may have persisted in the body fur of the animals or in the linings of their respiratory tracts, maintaining a state of autosensitization on repeated trials. This suggests that the asthmatic attacks observed were evidence of a state similar to status asthmaticus.

Although this remains a possibility we have called this response "learned asthma," since it meets the following criteria of conditioning outlined by Andrews:[1] (1) There must exist from the beginning a combination of an unconditioned stimulus and unconditioned response that is dependable, permanent, and not subject to extinction. (2) Then there must be paired with this combination in the proper fashion an extraneous neutral or conditioned stimulus (CS) that thereafter must be presented regularly along with the unconditioned stimulus and unconditioned response. The conditioned stimulus should not in itself induce the response. Our chamber served as the conditioned stimulus. After a sufficient number of trials pairing the conditioned stimulus with the unconditioned stimulus-response combination, the conditioned stimulus by itself must call forth the conditioned response. Four animals in the present study fulfilled these requirements. In addition, 5 of the 10 animals run in the smaller chamber also fulfilled these requirements. They had nine daily attacks in response to the CS.

The extinction of the conditioned reflex lends weight to the finding that this is a learned phenomenon. The question of whether the type of reaction we observed is generalized anxiety or fear also can be raised. In several animals we tried to evoke the response by loud noises, pain, and shock, without producing the pattern of asthmatic breathing we found in the experimental chamber.

The problem of which elements in the experimental situation act as the specific CS to produce the asthma is not clear. Our experimental chamber acted as the CS and could be viewed as a summation of multiple stimuli. It is possible also that internal changes within the animal acted as cues to the respiratory conditioning following the placing in the chamber. It should be noted that the conditioned responses were not as severe as the allergic responses. Such fractional reactions are commonly seen in the conditioning process.

The demonstration that asthma can be learned is of importance not only in relation to allergic phenomenon but also in relation to any agent, physical irritants, odors, or psychological factors, that may produce bronchiolar constriction. The learned responses reported herein bear a striking resemblance to human asthma as well as experimental allergic asthma. Whether the attacks reported are analogous to human bronchial asthma is still subject to much controversy. Asthma must be explained ultimately in terms of the basic pathophysiological change of bronchiolar constriction, which in turn produces the signs and symptoms.

Further studies in progress will enable

us to measure quantitatively the physical properties of the lungs as a direct index of bronchiolar function in experimental and conditioned asthma. This will permit a precise definition of asthma in terms of airway resistance and eliminate the dependence on clinical impression of animals' respiratory patterns. This procedure also has considerable importance for further psychological studies of learned asthma and should tell us whether learned asthma is physiologically similar to allergic asthma.

Summary

1. Young male guinea pigs previously made sensitive to egg white responded to a spray of homologous antigen with a respiratory syndrome that bears a marked clinical resemblance to human bronchial asthma.
2. A difference in susceptibility to attacks was found as well as a range of latency time in the onset of attacks in the animal population, suggesting inherent biological differences in disease reaction patterns.
3. Four of the 6 guinea pigs who completed 13 trials and 5 out of 10 in the later study group had learned asthmatic attacks that extinguished rapidly. The role of autosensitization, generalized fear, fractional responses, and learning have been discussed.
4. In studies in progress we are eliminating the dependence on clinical signs by measuring quantitatively airway resistance in the guinea pig. The airway resistance reflects the bronchiolar state and should indicate whether conditioned asthma is physiologically the same as experimental asthma.

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