FACTORS INFLUENCING ODOR SENSITIVITY IN THE DOG

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FACTORS INFLUENCING ODOR SENSITIVITY IN THE DOG

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This report covers an investigation of the possible influence of adaptation on the ability of dogs and human subjects to detect alpha-ionone; the form of the concentration-response function for alpha-ionone in dogs; the time trained dogs required to reach stable performance levels on low concentrations of alpha-ionone and its relation to adsorption time in olfactometers; the concentration response function for amyl acetate in the dog and quantitative measures of flow rate and associated parameters in dogs sniffing odor and air. The

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20. ABSTRACT Contd.

relevance of data to mechanisms of olfactory discrimination is discussed. Data on flow rate changes obtained from dogs not trained to detect odors presented is also given.

SUMMARY

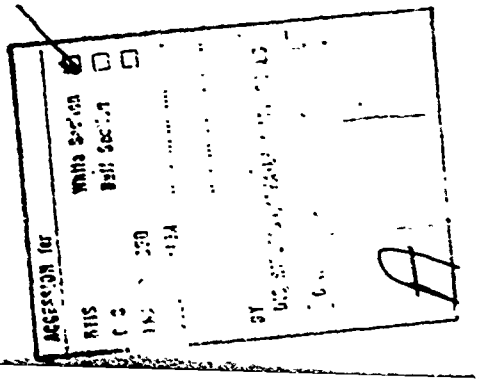
This report covers an investigation of the possible influence of adaptation on the ability of dogs and human subjects to detect α-ionone; the form of the concentration-response function for α-ionone in dogs; the time trained dogs required to reach stable performance levels on low concentrations of α-ionone and its relation to adsorption time in olfactometers; the concentration response function for amyl acetate in the dog and quantitative measures of flow rate and associated parameters in dogs sniffing odor and air.

The results of three different approaches demonstrate that adaptation to the test either does not occur or does not significantly influence quantitative measures of detection performance under the conditions of the expeirment. These approaches were: estimation of expsoure time to odorant during trials; examination of the effects on performance of varying intertrial intervals from 30-90 secs and analyses of frequency of errors following a correct response.

The concentration-response relation for α-ionone in three dogs consists of two limbs separated by a double reversal or notch which is statistically significant. The upper, slow ascending limb is best fitted by a parabolic function and the lower, steeply ascending limb is best fitted by a cubic function.

The concentration-response relation for amyl acetate in seven dogs reaches an asymptote at 10^{-5} of vapor saturation and descending towards a threshold below $10^{-6.5}$ of vapor saturation. The odor was presented in descending concentration series. The magnitude of the performance decrement at a given concentration was again found to be a function of the magnitude of the preceding concentration decrement. Variance in performance for most points was low as judged by a S.E. of less than $\pm$ 3% and provides no support for the view that age, sex or individual differences are important determinants of detection ability for all odors.

Quantitative analysis of duration flow rate, volume, and frequency of air and odor inhaled by a dog involved in an odor detection task reveal that mean duration of the sniff is relatively invarient at about 100 msec. The mean frequency of sniffing, however, increases at or below threshold concentration while the mean flow rate of the maximum sniff in a series declines with decreasing concentrations. The possible relevance of these data to mechanisms of olfactory discrimination is discussed. Data on flow rate changes obtained from dogs not trained to detect the odors presented is also given.



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PART I. ODOR DETECTION PERFORMANCE IN DOG AND MAN

Introduction

Our previous reports have described techniques and apparatus for investigating odor detection in dog and man and its use in determining concentration-response functions for α-ionone in both dogs and human subjects. These results demonstrated certain irregularities or notches in the concentration-response curves, and of the dogs superior detection abilities, and of factors influencing performance.

In this report we conclude the initial phase of this series of studies whose overall aims have been threefold: (1) to establish a technique for extracting reliable quantitative information on the dogs ability to detect odors, (2) to explore carefully (and in greater detail than has previously been attempted) certain of the performance and instrumental factors that might influence results obtained by this technique, and (3) to assess the potential of the concentration-response function as a probe for revealing events with a potential basis in receptor mechanisms.

This section of the present report covers the possible role of adaptation effects; the time required to reach maximum stable performances, and analyses of the concentration-response relations in dogs to determine what functions best describe them.

Some introduction to problems of adaptation is pertinent. Numerous studies have demonstrated that in human subjects the perceived intensity of an odor - even in low concentrations - decreases during prolonged exposure to that odor. The speed of this adaptation process and of recovery from adaptation varies from-odorant-to-odorant and increases with the intensity of the stimulus. No data of this kind exist for the dog. It must be assumed, however, that adaptation to α-ionone could occur in dogs and that it might therefore have depressed the observed performance. Reduced to experimental terms the question becomes: was the intertrial interval long enough to allow recovery from any adaptation to the test odor that might have occurred? However, some light on the possibility that the dogs adapted in the first place can also be drawn from knowledge of the exposure time. We have, therefore, used three approaches to evaluate this question: (1) observation of the dog's behavior during trials; (2) experiments involving systematic variation of the intertrial intervals; (3) analyses of previously obtained records to determine whether the probability of an error occuring was greater following a correct than an incorrect response.

Turning to another question this report also provides an analysis of previously obtained data for concentration-responses relations in dogs, aimed at determining (1) if the notches in the curves are statistically significant and (2) if so, do they define the transition between two segments of the curve fitting different functions. The significance of this analysis lies in the possibility (previously discussed) that the curve is the summation of two independent curves reflecting the action of two different mechanisms operating at the receptor level.

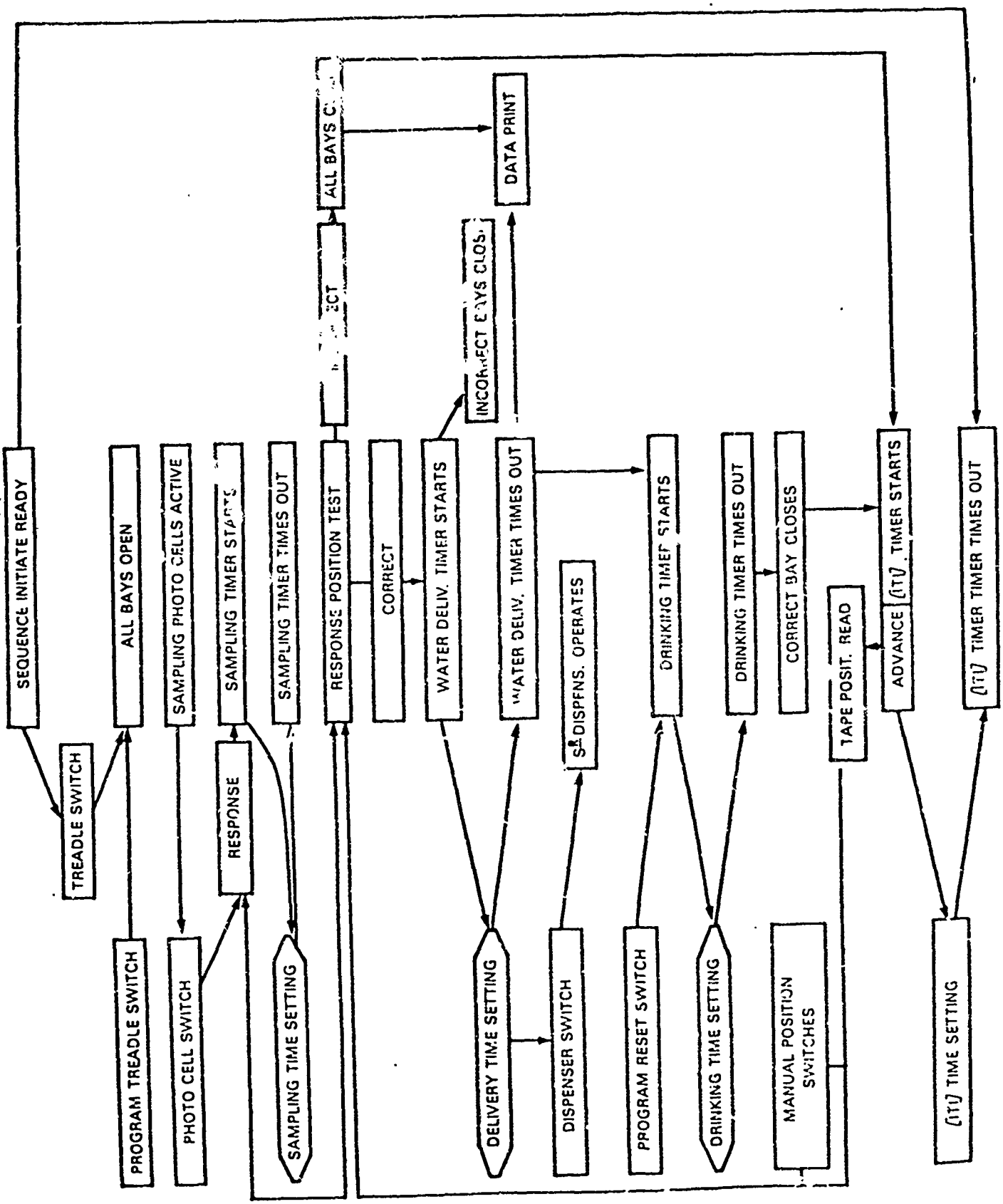


Fig. 1. Flow diagram for program controlling operation of test chambe . I.T I. = intertrial interval.

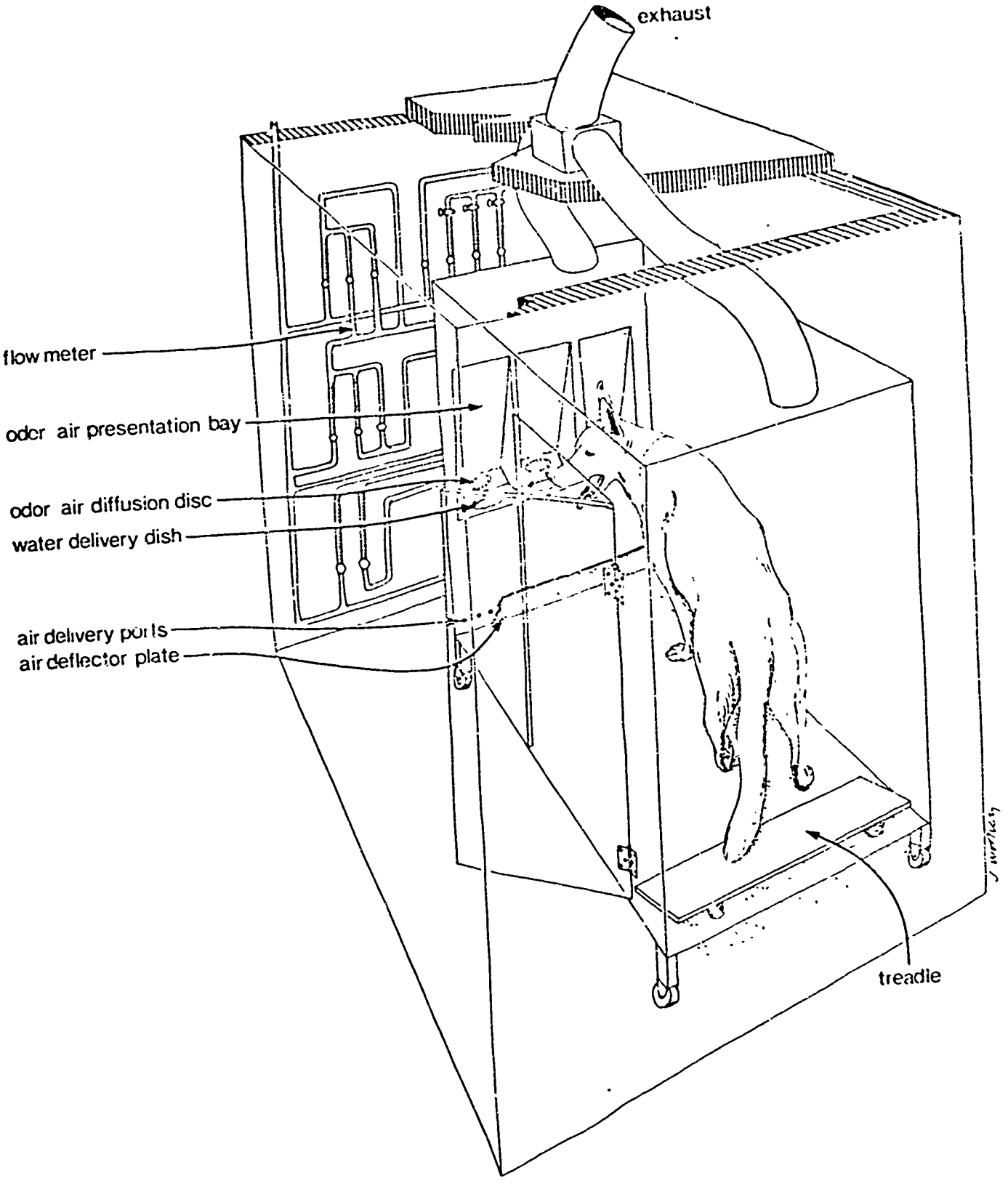


Fig. 2. Simplified view of controlled environment chamber.

Methods

The apparatus and methods used for training and testing both human and canine subjects have been outlined in previous reports. In brief, thirsty dogs are trained in a programmed odor-choice apparatus to sample each of three odor-air presentation bays and indicate (by the sustained interruption of a photocell beam) which of three bays is associated with an odor. If a correct choice is made the dog is rewarded with water delivered to a cup inside the bay. If incorrect, access to the bay is blocked. Each bay receives filtered air from an air-dilution olfactometer.

Details of the program controlling the apparatus are shown in Fig. 1 while Fig. 2 summarizes the main features of the test apparatus in a simplified semischematic form. The height of the room has been reduced and certain details omitted for the purpose of illustration. In particular, a gas chromatograph and water reservoir bottles normally rest on the roof of the chamber and air conditioning and air purification units are housed on the roof of the room. An observation window has also been omitted from the far wall of the room and the vapor saturator, temperature controlled bath and cold trap are hidden by the "chamber". The olfactometer is shown in highly simplified form. Although a dog is shown in the chamber it is large enough to accommodate a human subject.

The human subjects were tested in the same apparatus but rewarded 10^{c} each time a correct choice was made. In addition the subjects sniffed through a teflon-covered cone, one end of which was placed over the outlet of the vapor diffusion disc in the test bay.

The procedures we used to evaluate possible adaptation effects were of three kinds: (1) The maximum period of exposure to the test odor was estimated by watching the dog through the one-way glass panels at the front of the apparatus. The period the dog spent sampling blank air and odor bays was timed with a stop watch. We selected for observation blocks of ten trials chosen at random from each experimental session. In the case of human subjects each timed the duration of the sampling period with a stopwatch (the sampling period was defined as the time elapsing between the opening of the bay doors and the making of a choice. It did not include the intertrial interval. (2) A further approach was to analyze the sequence of correct and incorrect responses with the assumption that - if adaptation occurred - the increase in erros would be greater following the initial (as opposed to subsequent) exposures to odor in a block of trials. (The initial exposure in a block came after a period in which the dog had not been tested for more than an hour.) The data chosen were the first five trials in each of the final ten sessions for Dogs 1 and 4 (the best and poorest performers, respectively). This was repeated for each test concentration. (3) The performance of one dog was tested on longer, intertrial intervals. 100 trials were run, using $10^{-5.5}$ α-ionone, on each of the following intervals: 30, 45, 60 and 90 secs. The assumption was that if adaptation occurred (and the standard 30-sec. interval was too short to allow partial or full recovery) then increasing the intertrial

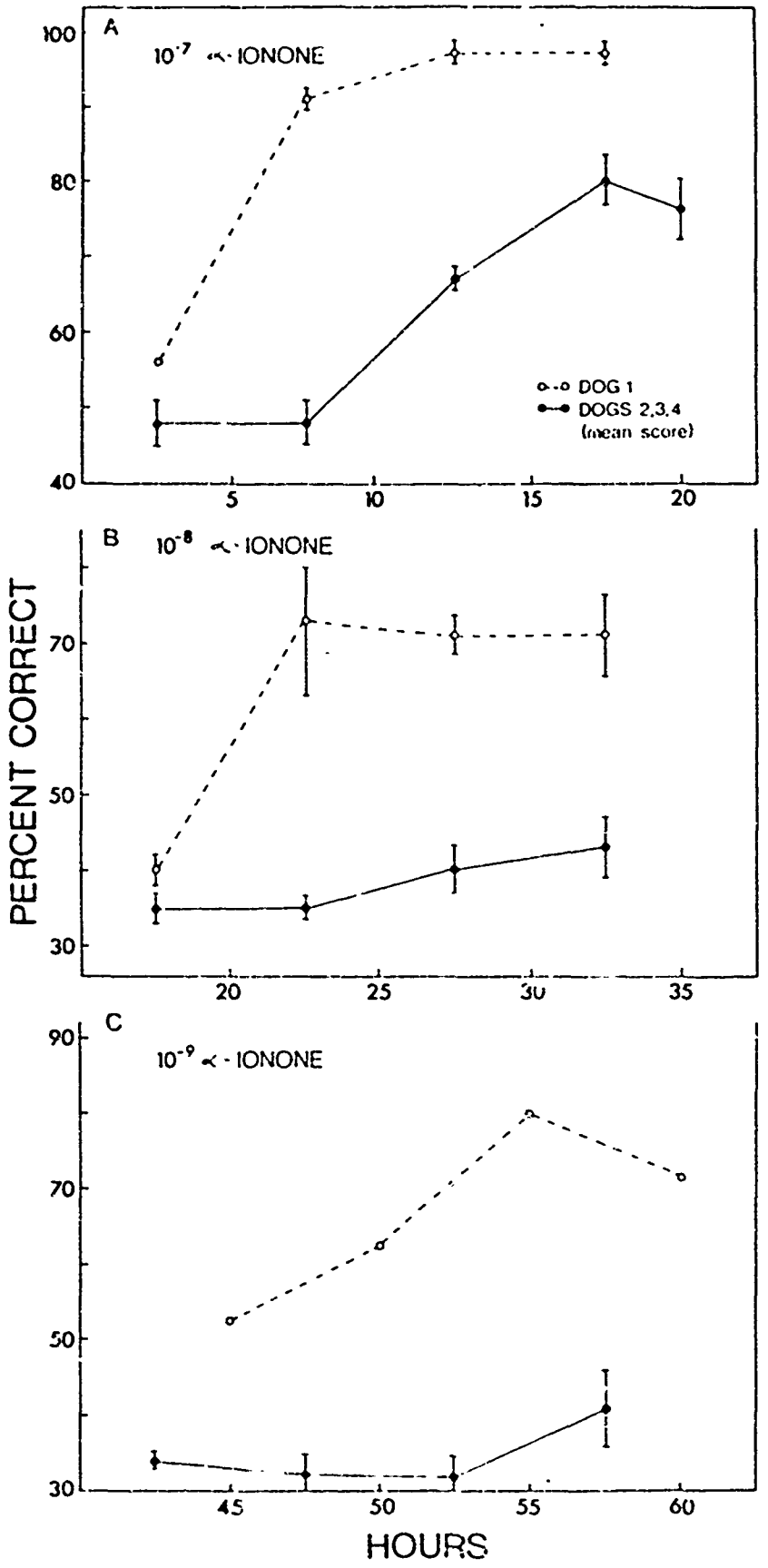


Fig. 3. Time required for dogs to reach maximum stable performance.

interval should permit at least some improvement in performance.

Turning to another problem data were obtained relating to the saturation time of air lines in the olfactometer. In three of the lowest concentrations of α-ionone tested (10^{-7}, 10^{-8}, 10^{-9}) the time required to reach a stable performance was determined. A parallel gas chromatographic evaluation was also made of time required to reach equilibrium concentrations when the output of the olfactometer was bled to the flame ionization detector.

Results

(a) Time required for performance to stabilize.

The results of the time required for dogs to reach a maximum stable performance are shown in Fig. 3. Because the performance of Dog 1 was considerably superior to all other dogs its scores are plotted separately. Each score is the average of at least one session (50 trials) ± S.E. Note that the ranges of both time and scores differ on each graph (chance level is 33%). Scores obtained before 18 hours in the middle graph and 43 hours in the bottom graph are not given since they were at the chance level.

The main point which emerges from these findings is that time to reach a maximum stable performance increases as a function of decreasing concentration. Gas chromatographic observations made with higher concentrations are consistent with the view that instrumental rather than training variables are mainly responsible for the long lag time in the stabilization of the dogs' performance. In other words, it may take up to 52 hours before the concentration delivered from the olfactometer at a setting of 10^{-9} of saturation actually reaches this value at the outlet of the olfactometer.

(b) Adaptation.

The results of the three procedures used to assess the possibility that adaptation could have reduced the performance levels of the dogs were as follows:

(i) Duration of exposure to odor. When fully trained, dogs seemed highly consistent both in patterns of approach and sampling from the presentation bays and in average duration of sampling. Sampling times for odor and blank air trials respectively averaged 1.04 and 0.98 secs based on analysis of 10-trial blocks selected at random from a number of sessions. However, at very low concentrations dogs made up to twice the number of discreet sniffs from each bay although their total sampling time remained relatively constant. In practice the 25-secs limit set on sampling was reached only when dogs ceased to perform.

Since a dog often sampled an odor bay more than once during a trial the total exposure time was greater than 1 sec. However, except at the lowest concentration, stimulus exposure due to active sampling rarely exceeded a total of 5 secs. This would be accumulated, for example, by

four or five one-second sniffs, each separated by one or two seconds of sampling blank air. In addition, in trials where a correct choice was made, there are two further periods of potential exposure time to odor. The first is imposed by the requirement that to indicate a choice the dog keep its snout in a bay for five seconds. This adds four seconds to the total. The second period is the time required to drink the reward. In the case of two dogs observed over several sessions this time was 3 - 4 seconds for each dogs.

Thus a conservative estimate for the maximum total adaptation time is 12 seconds per trial. The actual exposure time would be less since neither active sniffing not nasal inspiration during normal breathing occurs continuously throughout this period.

Human subjects varied from a few seconds to several minutes in the time they spent sampling bays before reaching a choice. At higher concentrations sampling was generally rapid. Near threshold, however, subjects would occasionally take up to 4 - 5 mins.

To determine whether longer sampling time was associated with a changed level of performance, correlation coefficients were calculated between mean sampling time and mean performance scores of all subjects for each concentration. The results show that no significant correlations exist. Thus there is no evidence that adaptation effects appear when exposure to the odorant is prolonged.

(ii) Analysis of sequence of correct and incorrect responses. Session records of trials run in the experiments that established the detection curves for alpha-ionone were analyzed to determine whether there were significantly more errors following the initial exposure to an odorant in a block of trials. Results of this analysis appear in Table 1. The data are given in 2 x 2 contingency tables showing frequencies of occurrence for combinations of correct ("1") and incorrect ("0") responses. Values of chi-squared with associated probabilities are given for several of the lower concentrations in 2A., and for each of the four sets of contingencies in 2B. It can be seen that the combination of correct response following a correct response occurs with a frequency significantly greater than that of other combinations. The two instances where this is not true are at 10^{-7} and 10^{-8} in A.; in these cases, no combinations occur significantly more often than others. Among the four combinations, the frequencies of an error following a correct response are generally the lowest. We therefore find no evidence of adaptation effects in so far as they are reflected in errors following correct responses. Further examination of response sequences beyond the fifth trial revealed no instances of repeated error runs separated by one or two correct responses, no alternating patterns of correct and incorrect responses. While runs of several errors are seen in some records, these are generally infrequent except for concentrations at or below threshold where chance-level performance occurs.

A.

10^{-3}

	1	0
1	10/10	0/0
0	0/0	0/0

10^{-4}

	1	0
1	10/9	0/1
0	0/0	0/0

10^{-5}

	1	0
	10/8	0/1
	0/1	0/0

10^{-6}

	1	0
1	7/8	0/0
0	3/2	0/0

	Second Trial	
First Trial	Dog 1/Dog 4	

χ^2= -/4.4; p = -/<.05

10^{-7}

	1	0
1	2/3	1/2
0	3/3	4/2

χ^2= 0/.42; p = .99/.5

10^{-8}

	1	0
1	2/1	2/2
0	4/4	2/3

χ^2= 1.4/1.0; p = .3/.2

B.

	Second Trial 1	Second Trial 0
First Trial 1	42/39	5/7
First Trial 0	15/14	8/10

χ^2= 4.5/4.7; p = <.05/<.05

	Third Trial 1	Third Trial 0
Second Trial 1	48/42	9/10
Second Trial 0	9/6	4/12

χ^2= 7.4/11.9; p = <.01/<.001

	Fourth Trial 1	Fourth Trial 0
Third Trial 1	51/38	6/10
Third Trial 0	7/8	6/14

χ^2= 7.1/10.4; p = <.01/<.005

	Fifth Trial 1	Fifth Trial 0
Fourth Trial 1	57/36	1/11
Fourth Trial 0	6/9	6/15

χ^2= 20.7/8.7; p = <.001/<.01

Table 1. Analysis of response sequences for Dogs 1 and 4: Correct ("1") versus error ("0") responses using last 10 sessions per concentration. A: First trial paired against second trial. B: Sequential pairings of adjacent trials based on total data for 7 test concentrations, 10^{-3} through 10^{-9}.

Table 2. Comparisons of session socres for standard and increased length inter-trial-intervals.[1]

A. Data from Dog 4 at a concentration of $10^{-5.5}$.

Standard (30-sec.) ITI	Increased inter-trial intervals	
100	96	45 sec.
88	97	
96	92	60 sec.
98	90	
91	93	75 sec.
$\bar{X}$ = 94.6	$\bar{X}$ = 93.6	

B. Data from two human subjects at a concentration of $10^{-4.5}$.

Standard (30-sec.) ITI	ITI increased to 90 sec.
80	87
100	80
70	73
80	73
47	50
67	80
67	47
80	80
$\bar{X}$ = 73.9	$\bar{X}$ = 71.3

[1] Performance scores are given as percentages based on 50-trial sessions.

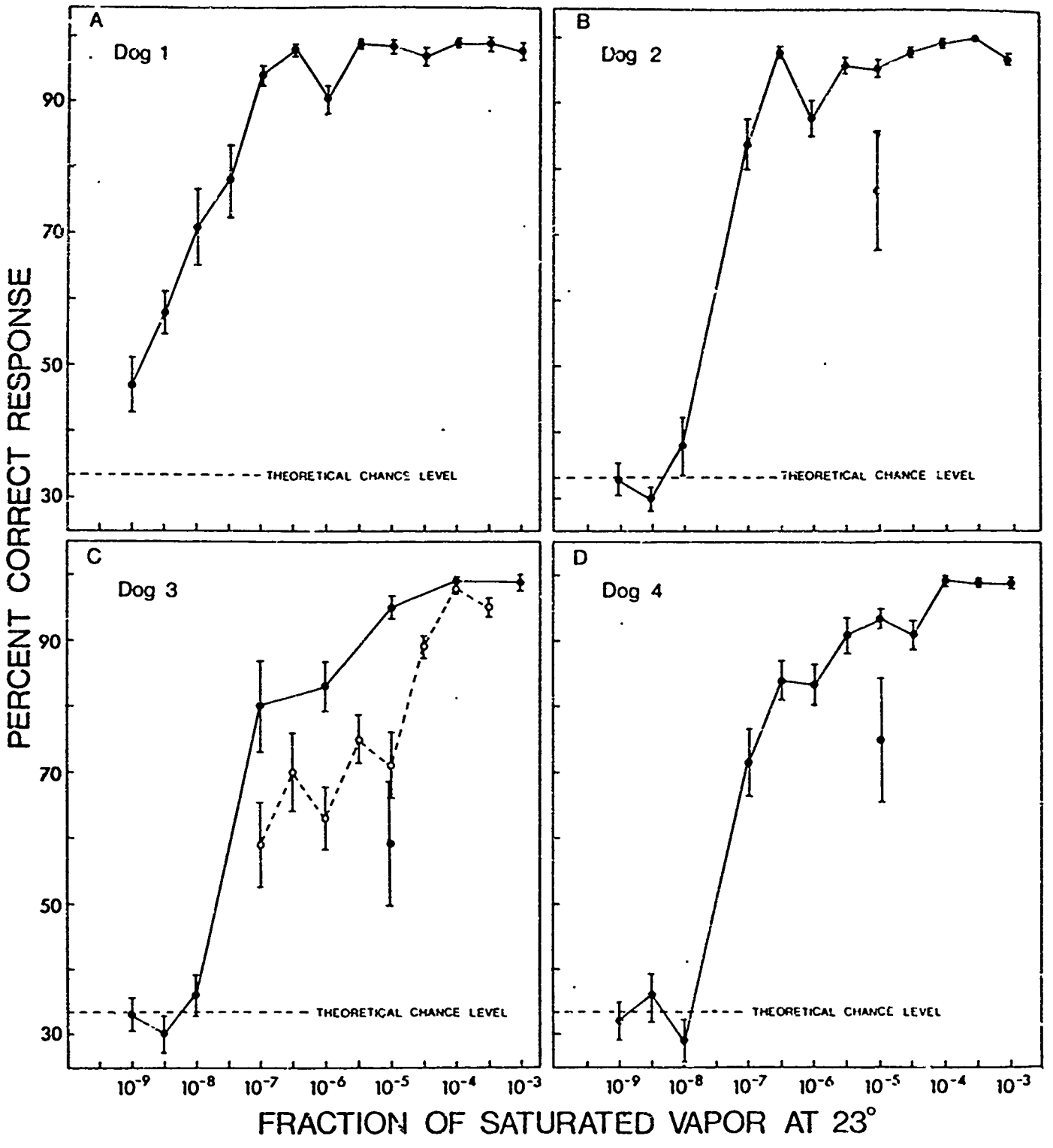


Fig. 4. Concentration-response curves for α-ionone in four dogs.

(iii) Variation of intertial interval. The effects of increasing the intertrial interval on the performance of one dog and two human subjects are summarized in Table 2. Mean values for the series of intertrial intervals show only slight effects of increased intertrial length on performance and they are in the opposite direction from that expected if adaptation were occurring. Probabilities of the outcomes in A and B (Table 2) are $p = 0.42$ and $p = 0.29$ respectively (Mann-Whitney U-test).

(iv) Correspondance of performances on ascending and descending series. To the extent that adaptation might be expected to increase variance the close correspondance between the performances of Dog 1 on ascending and descending series is significant (Fig. 4).

(c) Analysis of stimulus-response functions for α-ionone.

The data obtained during previous concentration series with four dogs are shown in Fig. 4. Curves for Dogs 1, 2 and 4 are constructed from scores averaged overall test series and include scores on the ascending series (Dog 1 only). Dog 3's performances on the two descending series diverged so widely that they have been plotted separately. Each point on these curves as well as those for the ascending series of Dog 1 ($10^{-8.5}$ and $10^{-7.5}$) is the mean of a minimum of 500-750 trials $\pm$ S.E. Each of all the remaining points is the mean of 1000-1500 trials $\pm$ S.E. (The isolated point at 10^{-5} is the score attained when a two-log decrement in concentration was introduced initially.)

These curves are shown to demonstrate the marked discontinuity in slope seen in all these curves centered on 10^{-6}. As previously discussed, this discontinuity or actual notch in the curve marks the division of the curve into two segments. If this is assumed, then the upper and lower limbs are displaced relative to one another on the concentration axis by about one-half log unit. If we focus attention on the notch or discontinuity it is possible to assess its statistical significance by analyzing the differences between the mean score at the base of the notch with score at the next highest and lowest concentrations ($10^{-5.5}$ and $10^{-6.5}$). When this is done (Table 3) both comparisons show significant differences for Dogs 1 and 2.

	Dog No.			
	1	2	3	4
$10^{-5.5} > 10^{-6}$	.016	.028	.048	.075
$10^{-6} < 10^{-6.5}$	.048	.016	.155	.274

Table 3. Probabilities associated with performance differences at three test concentrations. (Significance levels are for the Mann-Whitney U test applied to data from the final five sessions at each test concentration.)

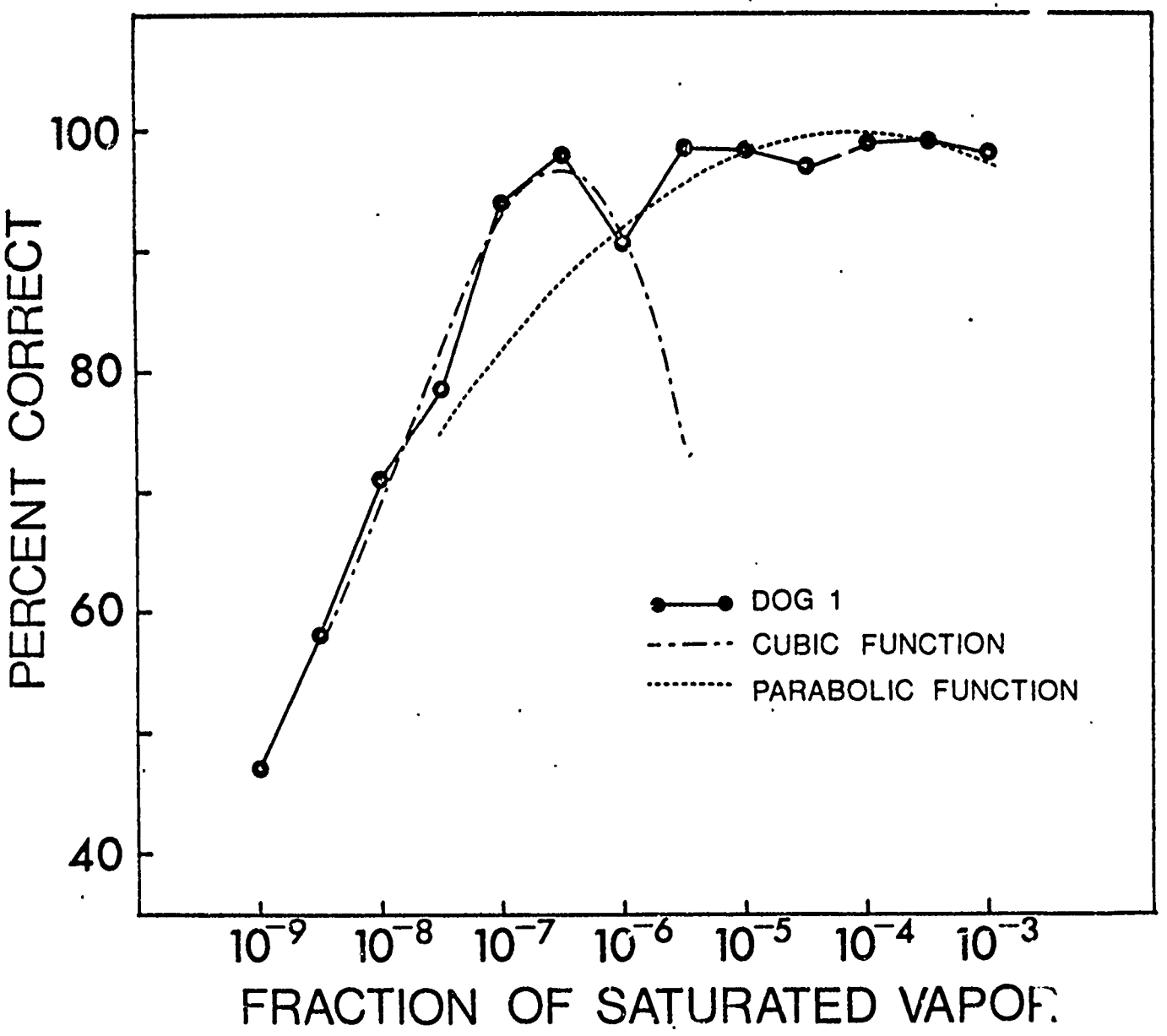


Fig. 5. Least square fits to performance scores of Dog. 1.

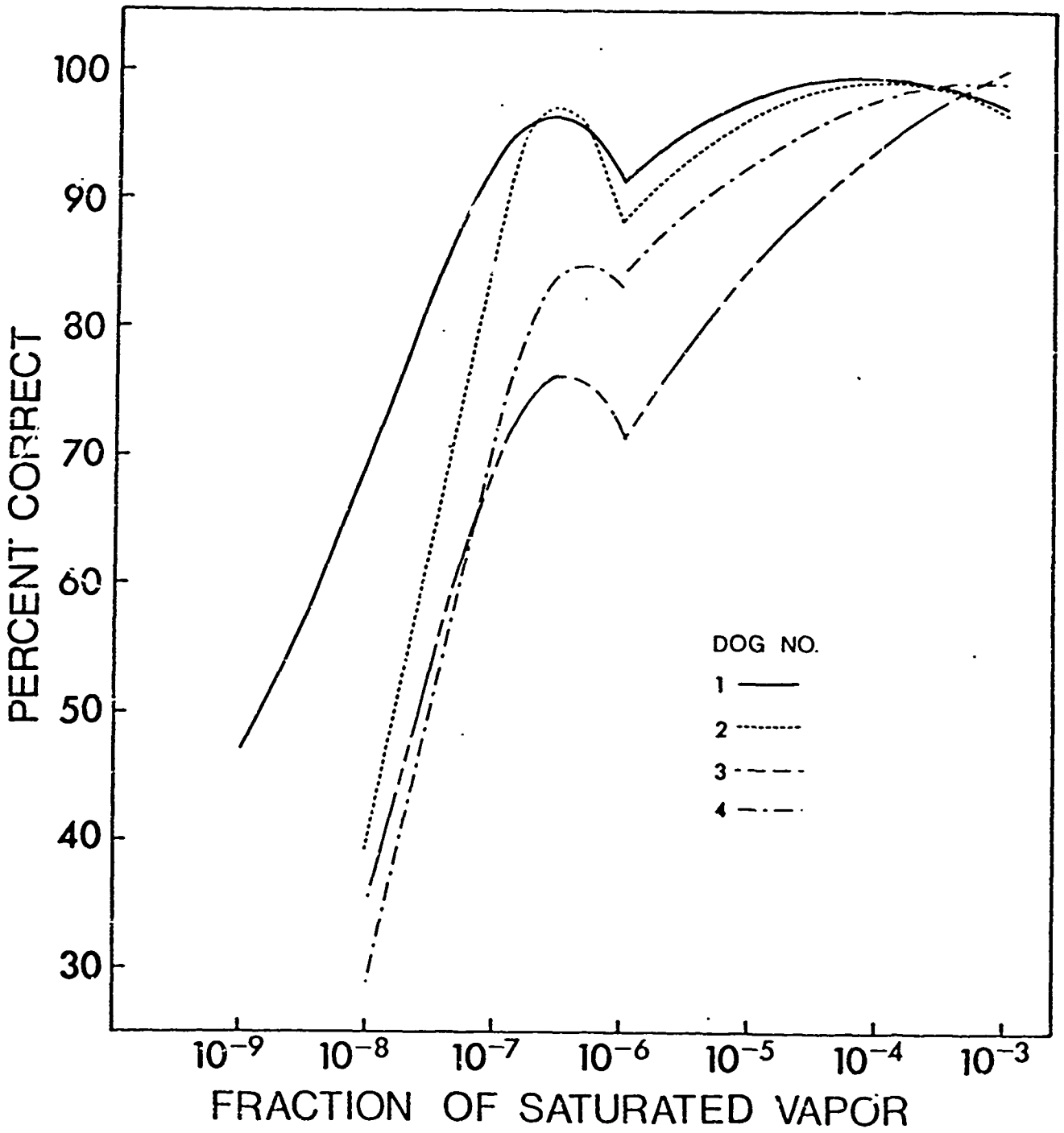


Fig. 6. Least square fits to performance scores of four dogs tested on α-ionone.

For Dog 3 only one comparison is significant as for Dog 4 neither are. It appears, then, the phenomenon is sufficiently real to justify further analysis on the assumption that a different function can be fitted to each limb of the stimulus-response curve. This has been done in Fig. 5 for Dog 1's data. Least square curve fits were calculated. The best fit to the lower limb is a cubic function, while the upper limb more closely approximates a parabolic function. In Fig. 6 the analysis has been extended to all four dogs.

Discussion

(a) Adaptation. We assumed that adaptation to the test odor might occur and that it might therefore have depressed performance. The results and analyses of the present study do not eliminate the first assumption but provide no evidence for the second. In the case of dogs the potential exposure time to an odor during a trial was in any case very brief (< 12 secs.). In both canine and human subjects analysis of order effects failed to reveal any increase in errors following the initial trial of a series. Finally, increasing intertial intervals does not lead to any significant improvement in performance. It thus seems reasonable to conclude that the notch in the stimulus-response function, the form of these functions and the threshold magnitudes found were not influenced by adaptation to the test odor.

(b) Time required to reach stable performance. We concluded that the time required for performance to stabilize increases as concentration decreases. In particular, performance at 10^{-9} α-ionone took over 50 hours to stabilize. It might be assumed that these are primarily training effects. While this cannot be eliminated as a contributing factor the evidence from gas chromatography makes it clear that sorption of α-ionone on the walls of the olfactometer and delivery lines significantly prolong the time required to reach quilibrium conditions. At the lower concentrations this effect could well account for the delay in reaching stable performance. A further point tending to confirm this assumption is that all dogs were highly trained and repeated this delay when retested on the same concentration at a later date.

(c) Concentration-response function. The analyses presented above clearly demonstrate the dual nature of the detection curves for α-ionone and establish the statistical significance of the notch separating the two components. The further conclusion that the upper limb follows a parabolic and the lower a cubic function strengthens the view that the curve may reflect the action of two distinct mechanisms at the receptor level. Since the concentration-response function for the frog has been shown to follow a hyperbolic curve (as would be predicted from an assumption that transducer events are described by the law of Mass Action), this evidence is consistent with the possibility that at least one component of the curve may preserve relations established at the receptor level.

II. ENHANCEMENT OF RESPONSE TO ODORS

Introduction

The previous annual report (1974) outlined results of a pilot study which showed that oral administration of 1 ml α-ionone to a dog markedly altered the dog's ability to detect this odor in the vapor phase. The odor was presented in a concentration of $10^{-4.5}$ of vapor saturation at which level the dog attained a performance-control trial of .89 ± 3.2%. In the two weeks following the ingestion the dog's performance oscillated mainly above baseline (89%) to reach a maximum level of 100% attained during the 7th and 8th days before returning to near baseline. This level exceeded that ever previously achieved by this dog on this concentration.

A further series of more extensive studies on this phenomenon are now in progress. To determine the specificity of the effect dogs are being trained on amyl acetate.

Definitive trials on this aspect will begin when a concentration of amyl acetate is reached that stabilizes performance in the 65-75% range. (This allows both declines and rises in performance levels follwoing odor and ingestion to be detected.) The information already generated, however, is pertinent to the question of whether the magnitude of concentration decrement preceding attainment of a stable performance determines the magnitude of the decline in this performance; to understanding the form of the stimulus-response function for olfaction and to the assumption that odor detection is markedly influenced by age.

Methods

3 German shepherds used in the previous study with α-ionone (2 female and 1 male) together with 4 younger German shepherds (2 male and 1 female) were trained and tested on the apparatus and according to the methods outlined in Part I, and in previous reports. When dogs attained a stable performance level at 10^{-4} concentration was lowered by one log step and they were retrained and tested. Concentration was then raised by one-half log unit, the dogs retrained and tested on this performance before returning to 10^{-5} once more. Thus two performance scores were obtained for 10^{-5}: one following a log unit drop in concentration and one following a half-log unit drop in concentration.

The odorant was obtained from Eastman Kodak Company and was purified by chromatography.

Results

The dogs' performance on 10^{-3} - 10^{-6} of saturated amyl acetate vapor is shown in Fig. 7. Data obtained subsequent to the completion of this graph show that the performance is still well above threshold at $10^{-6.5}$ and it appears probably (by extrapolation) that thresholds lie within the range 10^{-7} - 10^{-9} of vapor saturation. The notch in the curve at $10^{-4.5}$ is not statistically significant.

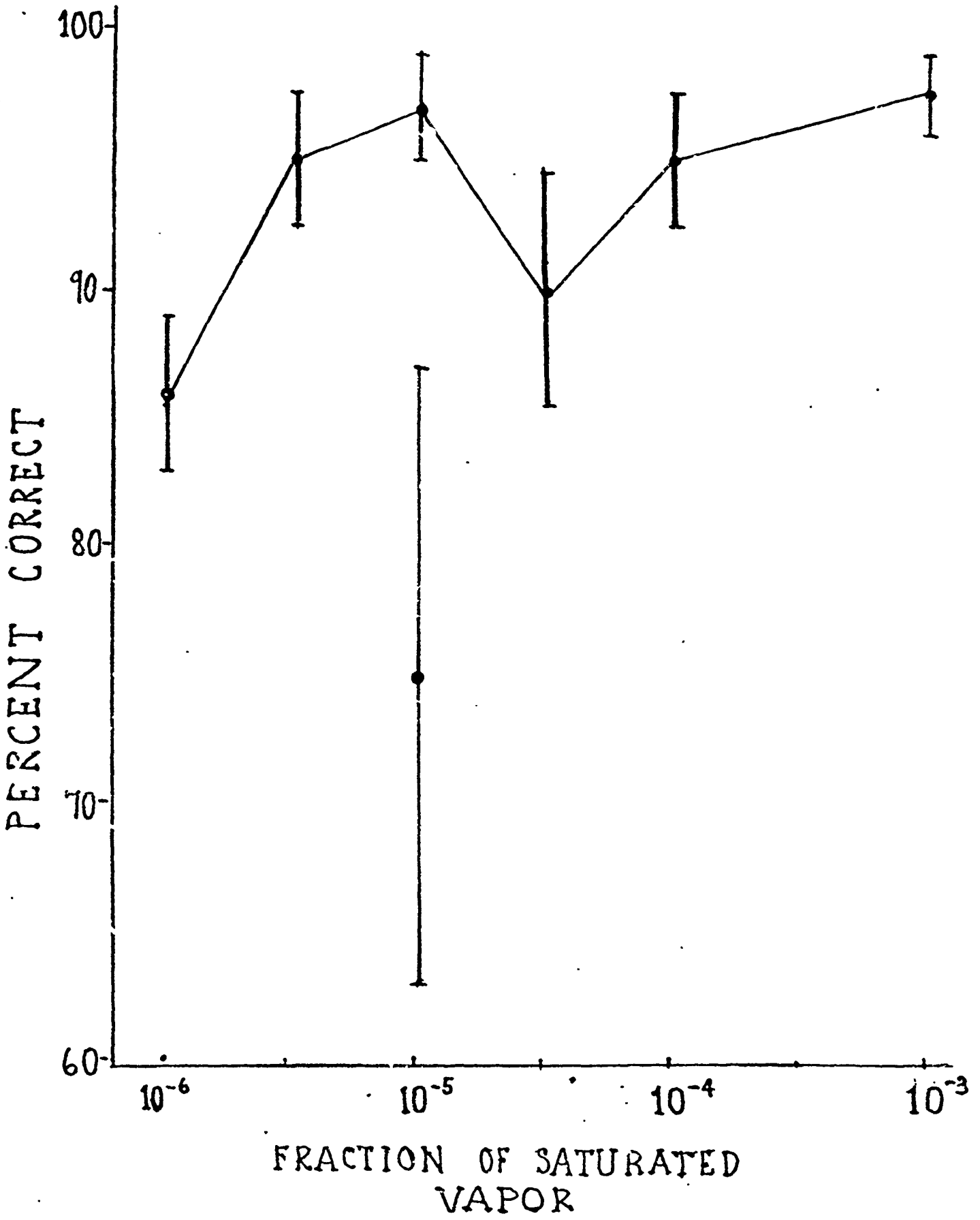


Fig. 7. Concentration response relations for n-amyl acetate. Each point is the mean ± S.E. of 1,400 trials.

The lower of the two points shown at 10^{-5} was obtained following testing at 10^{-4}. The upper point was obtained following retraining and testing at $10^{-4.5}$. Despite an adequate number of testing sessions performance remained highly variable in the first case. In the second case variance was markedly reduce while performance levels were considerably higher.

Discussion

Previously published thresholds for amyl acetate in man and rat are about 10^{-6} of vapor saturation. The present evidence suggests that the dog is superior to all species for which data currently exist having a projected threshold lower than 10^{-7} and possibly as low as 10^{-9} for some dogs. This corresponds to a superiority of some 10 - 1,000 over man and rats which is thus somewhat less than the dog's superiority over man for α-ionone (see previous reports). However, fuller discussion of this point is premature and must await completion of the present study.

The marked decrement in performance which follows a log unit drop in concentration as compared with that which follows a half log unit drop is consistent with the conclusions reached in studies with α-ionone. It provides further evidence that the level of performance attained following a decrement in concentration is a function of the magnitude of that decrement.

Despite the fact that 7 dogs were used in the present study the variance in performance among them is remarkably small being $< \pm$ 3% S.E. for most points. This suggest that neither sex, age, nor individual differences significantly influence performance at these concentration levels since age differences were in the order of 3 years and both sexes were represented. In contrast, the data for α-ionone showed a wider spread in scores between individuals.

III. RATES, AMPLITUDES AND PATTERNS OF AIR AND ODOR FLOW DURING SNIFFING IN DOGS

1) Measurement of sniff parameters in dogs performing an odor detection task

Introduction

Several lines of evidence point to the need for an accurate quantitative analysis of the characteristics of sniff cycles when an animal is actively engaged in an odor detection task. No such analysis has been made in any animal including man except in relation to isolated measures of sniff volume (1-3 l/min in man: D. Laing, personal communication, 1975) and sniff frequencies in rats (1-11 cycles/sec). These studies, however, provide no accurate information about the detailed composition of the sniff cycle or the extent to which it varies with concentration (particularly at low dilutions). We have therefore sought to provide such information for a dog performing a learned odor detection task. A dog was trained to respond

differentially to the presence of an odor in such a way that it sniffs through a pneumotachometer. The ultimate aim is not only to measure accurately the flow amplitude, duration and frequency but also to determine whether these parameters vary as a function of concentration, nature of the odor and of the task (i.e., whether it is odor discrimination or detection). If the mode of dispersal of odors within the olfactory organ is important (for example, in discrimination rather than detection) it should be reflected in such data. Since the work is still in progress and data have not yet been analyzed this is a preliminary report.

Methods

Subjects

Two female and one male German shepherd were used. One female was about three years old at the start of the experiments while the other dogs were about one year old. They were housed in temperature controlled indoor runways, fed laboratory chow ad lib, and placed on a 23-hour water deprivation schedule. During testing and training they received an average of about 400-600 cc of water as rewards. The difference between this quantity and 1500 cc was given to them early each morning following the day of testing.

Behavioral test apparatus

The apparatus provides two bays, one associated with the odor of amyl acetate, and the other a blank. The two bays are set in a wooden console. Two swinging metal doors carry the sniffing ports. They are counterweighted to allow the dog to push them open but can be latched in position to block the dog's access to the water bowls (visible beneath the doors). The experimenter releases the latch by remote control when the dog makes a correct choice. The bowls are gravity fed from calibrated water reservoirs in the upper section of the console.

Behind each sniffing port are two metal cylinders of similar length and diameter extended inwards by polyethylene cylinder. One of these is the Fleish pneumotachograph, the other is a dummy. To equalize flow resistance in the two cylinders the internal lumen of the dummy is fitted with a smaller cylinder. Since their relative positions can provide no differential cues, the cylinders occupy the same positions permanently. The cylinders are open at both ends but near the opening into the interior of the console there is a port on the floor of each polyethylene cylinder. This is made to accommodate a 10-cc vial, set so that its mouth is flush with the lumen of the cylinder. A loose glass wool plug about 3 cc in volume is placed into each vial. 25 drops of pentyl acetate in the diluent (ethylene glycol) are delivered to one vial and 25 drops of the diluent alone are delivered to the other. The sample vials are recharged after several runs are made to ensure reliable stimulus production. The relative positions of the vials (test and "blank") are varied according to a randomly determined sequence. Between trials each chamber was

flushed out with a fan to ensure that no odor would remain to interfere with the next trial.

Each of the sniffing ports is surrounded with a ring of foam rubber. This allows the dog to insert its snout into the port without irritation yet seals tightly enough to prevent air leaking around the dog's snout.

Odorant and concentration determination

N-pentyl acetate was chosen as the first odorant because it has previously been used in olfactory studies (on rats, rabbits, tortoises, pigeons and man) involving both electrophysiological and behavioral apparatus; has a sharp distinctive odor with a known trigeminal threshold lying well above the olfactory threshold and has no known biological significance for the dog. It was diluted with ethylene glycol (Baker reagent grade) to the appropriate concentration.

Since the odor in the cup is being diluted with the air drawn into the nosecone, the dog experiences a stimulus concentration considerably less than that present in the sample cup headspace. In order to estimate this dilution effect, a sample cup of saturated amyl acetate was put in place, air was drawn through the chamber as in sniffing, and then a sample was withdrawn from the chamber by gas-tight syringe and injected into the gas chromatograph (GC). Comparison of GC peak areas of these samples with equal volume samples of saturated amyl acetate vapor from the bottle headspace revealed that a dilution of about 1000 to 1 was being made from sample cup to pneumotach nosecone.

To avoid any confusion stemming from these differences we will refer to concentration in solution as % concentration and concentration in air as a fraction of vapor concentration (10^{-3}, 10^{-4} etc.).

Recording apparatus and its calibration

The output of the Fleish pneumotachograph was fed through a pressure transducer, amplified, monitored on an oscilloscope and stored on a tape recorder. Visual records of these tapes were later obtained from a multi-channel pen recorder. The pneumotachometer was calibrated by attaching a 1000-cc syringe and drawing air through it. The amplitude of the pen deflection was then plotted against flow rate. As can be seen in Fig. 5, the response is linear over a wide flow range under the conditions of the experiment.

Training and Testing

Summarizing information given in earlier reports: dogs were first trained to indicate which door was associated with odor by pressing on it with their snout, and later by inserting their nose into the nose cone. No limits were set on the response time. Dogs were tested on a descending

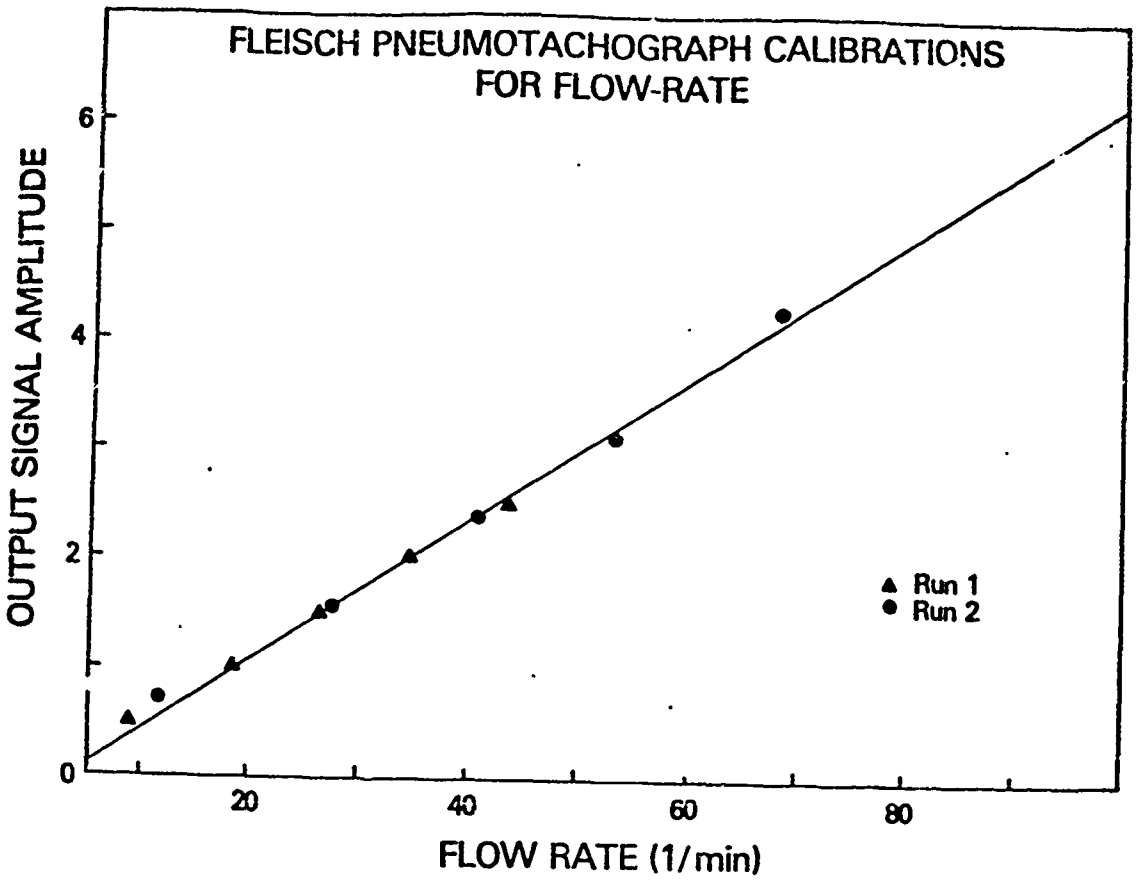


Fig. 8. Calibration curve for Fleish pneumatochograph. Amplitude of pen recorder (ordinate) is given in arbitrary units.

concentration series and responses to both odor and air were recorded. All concentrations, however, except the lowest, elicited performance scores well above the chance level.

Analysis of data

Since no previous study of this kind appears to have been attempted, analyses were directed towards identifying significant features in sniff patterns and determining how these differed (if at all) between air and odor for different concentrations of odors. Parameters chosen for initial study were: mean flow rate and volume of maximum sniff; mean duration of sniffs and of bouts; the number of sniffs in a bout and the number of bouts in a train. Sniff-volume was estimated by calculating the area of the equilateral triangle that best fitted the sniff trace. Since individual sniff records generally approximated a triangle the error involved is small.

Results

Table 3 summarizes the data obtained for two dogs over a series of concentrations of amyl acetate and selected relations are plotted in Figs. 9-11 for dog 2P. Since Dog 6S has only recently been trained and the data are limited little emphasis can be placed on them at this stage. However, on one point both dogs show a remarkably close agreement: the mean duration of the sniff is 10-11 for both dogs.

The data are expressed with reference to three basic categories of measurement: sniffs, bout and trains. We can define these as follows: a sniff is the smallest significant inhalation; a bout is a succession of sniffs not interrupted by removal of the dog's muzzle from the nose cone and a train is a succession of bout made at one choice point during a single trial.

The actual number of sniffs per bout ranged from 1-6 but the means for each pair of values (odor vs. air) obtained at each concentration was 2.3-5.1.

Sniff flow rates varied in amplitude with peak flows tending to occur later in a bout. The maximum was the penultimate or occasionally the final sniff for Dog 2P and appeared to indicate that the dog had reached a decision. This pattern has not yet developed in the data for Dog 6S. For Dog 2P the mean flow rate of these maximum amplitude sniffs was higher for odor than for air at all but one concentration (Fig. 9) but the limited data for Dog 6S do not show this. The differences are most significant for the highest concentration. Odor sniff amplitudes decline with decreasing concentrations without any evident change in the Standard Error of the mean. On the other hand, when the mean duration of the maximum sniffs in a bout is considered differences between magnitudes for odor and air are less evident. Means of maximum sniffs ranged from 41.4-72.5 ./min and from 66.3-78.0 cm^2 for air and different concentrations of odor.

DOG 2P

CONC.	NO. OBSERVATIONS		$\bar{X}$ NO. BOUTS PER TRAIN		$\bar{X}$ NO. SNIFFS PER BOUT		$\bar{X}$ BOUT DURATION (secs)		$\bar{X}$ FLOW RATE OF MAXIMUM SNIFF (1/min)		$\bar{X}$ VOL. OF MAX. SNIFF (cm^2)		$\bar{X}$ DURATION OF SNIFF (sec)	
	ODOR	AIR	ODOR	AIR	ODOR	AIR	ODOR	AIR	ODOR	AIR	ODOR	AIR	ODOR	AIR
1.0	9	8	2.1	1.0	3.0	4.1	.77	.76	72.5	58.4	68.8	69.4	.11	.10
0.1	5	4	2.6	2.0	2.4	3.0	.64	.55	66.1	57.8	71.4	75.5	.10	.16
0.01	12	11	2.4	1.9	3.1	3.1	.85	.70	60.5	59.5	78.0	71.4	.11	.11
.001	7	3	3.1	1.7	2.9	2.3	.72	.63	51.6	51.7	69.3	67.5	.09	.09
.001	5	5	1.8	2.2	5.1	3.6	1.26	1.02	48.0	41.4	66.4	66.3	.10	.11
	38	31												
$\bar{X}$			2.4	1.8	3.3	3.2	.85	73	59.7	53.8	70.8	70.0	.10	.11
DOG 6S														
1.0														
.01	5	3	1.4	2.1	2.4	2.2	.40	.59	39.5	44.0	38.6	41.3	.10	.11
.001														

Table 3. Summary of measurements of sniff parameters for two dogs trained to detect n-amyl acetate. The concentrations of n-amyl acetate are expressed at percent liquid dilution steps and are not the concentrations in the vapor phase. For Dog 6S data for 3 concentrations are combined.

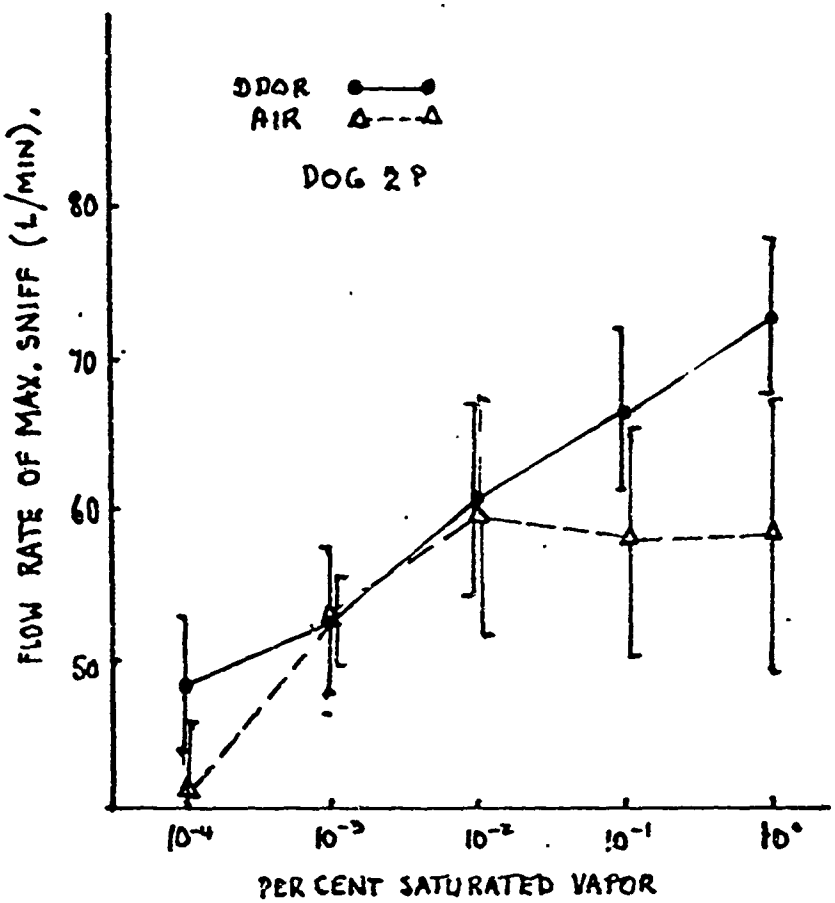


Fig. 9. Relation of mean flow rate of maximum amplitude sniff in a bout to concentration of n-amyl acetate.

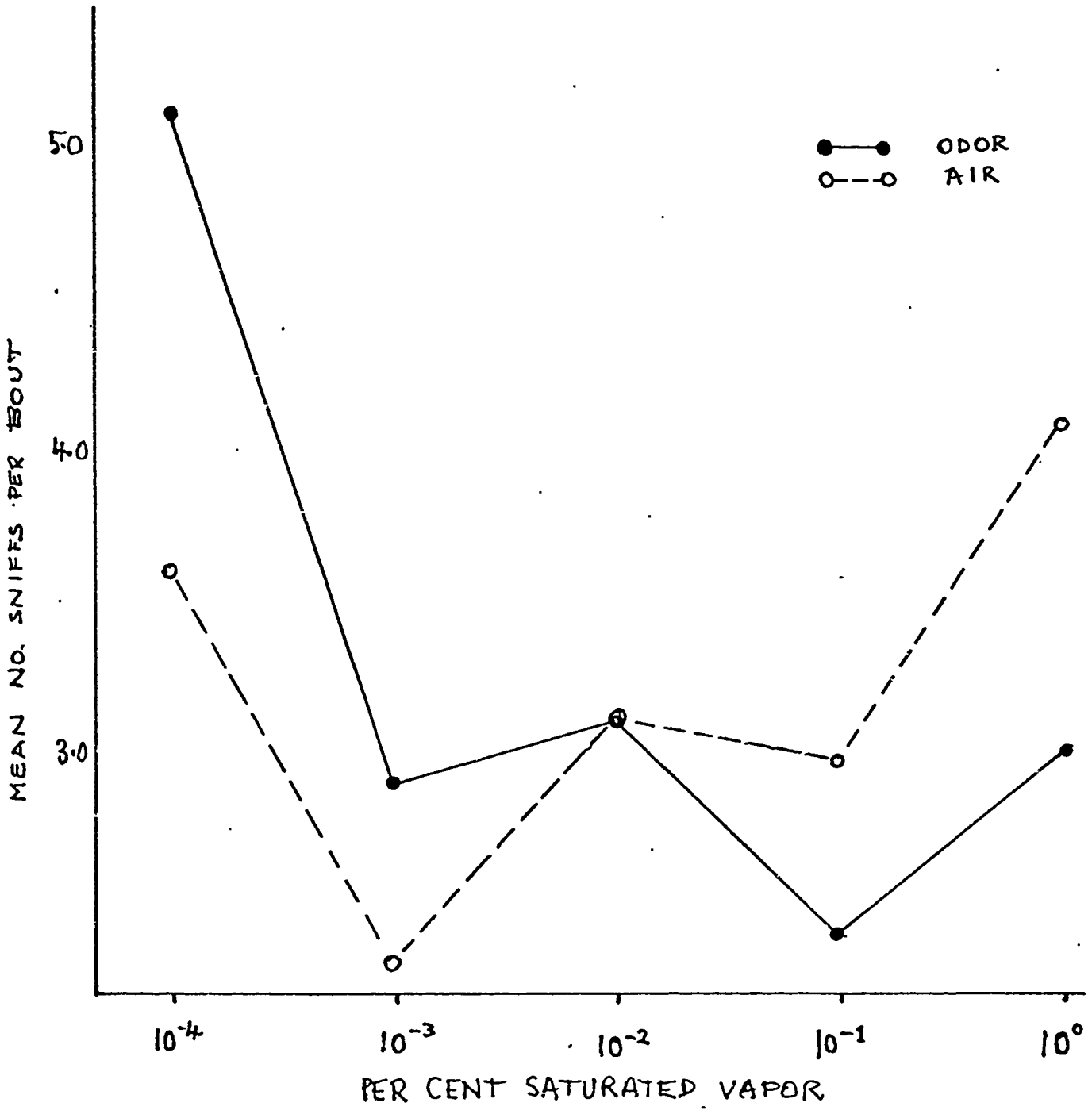


Fig. 10. Relation between mean number of sniffs per bout to concentration of n-amyl acetate.

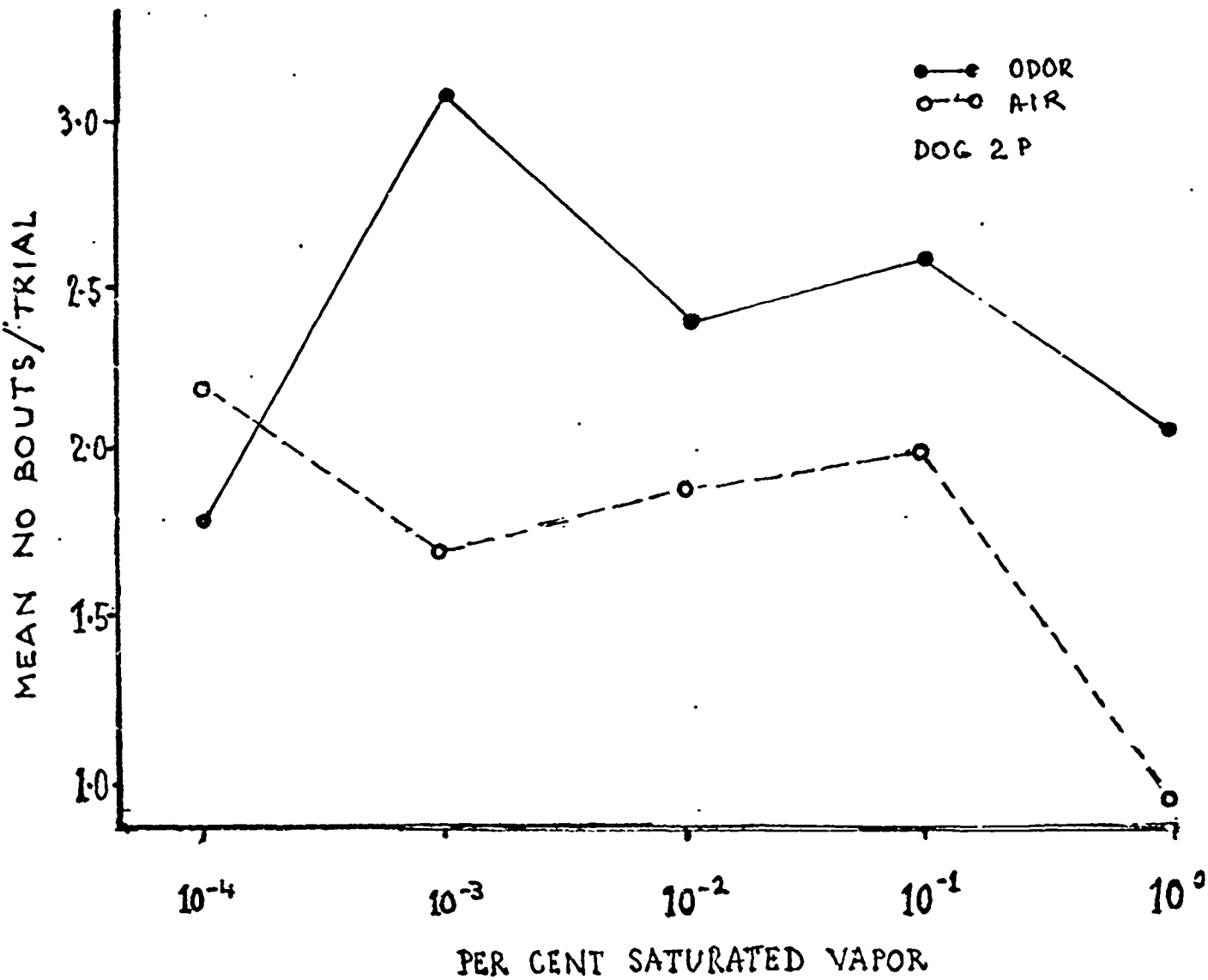


Fig. 11. Relation between mean number of bouts per train to concentration of n-amyl acetate.

Differences between sniff responses to odor and air are also evident in Figs. 10 and 11 . Fig. 10 shows a tendency for the mean number of sniffs per bout to be higher in relation to air than to odor while the opposite trend occurred at lower concentrations. From Fig. 11, however, it is clear that for all but the lowest concentrations tested the mean number of bouts per train is higher in relation to odor than in relation to air.

DISCUSSION

One unexpected and striking finding was the relative constancy in the mean duration of the sniff (150 msec). Differences are insignificant irrespective of the comparison (odor vs. air; different concentration of the odor or different in individuals) despite the wide variations in individual sniffs. The only exception may lie in comparisons between earlier and later sniffs in a bout. (Although data are insufficient to establish the point they suggest a tendency towards growth in sniff duration within a bout.) If this has any bearing on the mechanisms of olfactory discrimination it may indicate that there is an optimal time needed to disperse a "packet" of molecules across the receptor sheet. Beyond this, such factors as adaptation, or a decline in the efficiency of separation of molecules by sorption, may occur. Consistent with this interpretation the absence of differences suggests that sensory performance near threshold cannot be improved by changing sniff duration.

If sniff duration is not a significant variable the number of sniffs in a bout probably is. At threshold levels where performance is at or near chance frequencies increase markedly. Although it will require further data from other individuals to establish the generality of this effect it suggests that the dog sniffs more frequently rather than with more sustained sniffs when it is necessary to maximize detection. It is not yet clear whether there are any meaningful differences in the number of bouts per trial between odor and air and at different concentrations.

The clearest trend emerging from this study is in the decline in mean flow rate of the maximum amplitude sniff as a function of decreasing concentration. Again this is somewhat unexpected since it might be supposed that as the difficulty of the task increased the need to pull in more molecules per sniff might also rise. That is does not suggests that too many molecules at once may increase the difficulty of detecting a meaningful signal possibly because a higher proportion of them are likely to be contaminants (at least under normal environmental conditions). At higher concentrations the ease of the task makes volume an unimportant variable and high volume sniffs are as effective as low volume sniffs except when sniffing at the air port where lower volume sniffs become necessary.

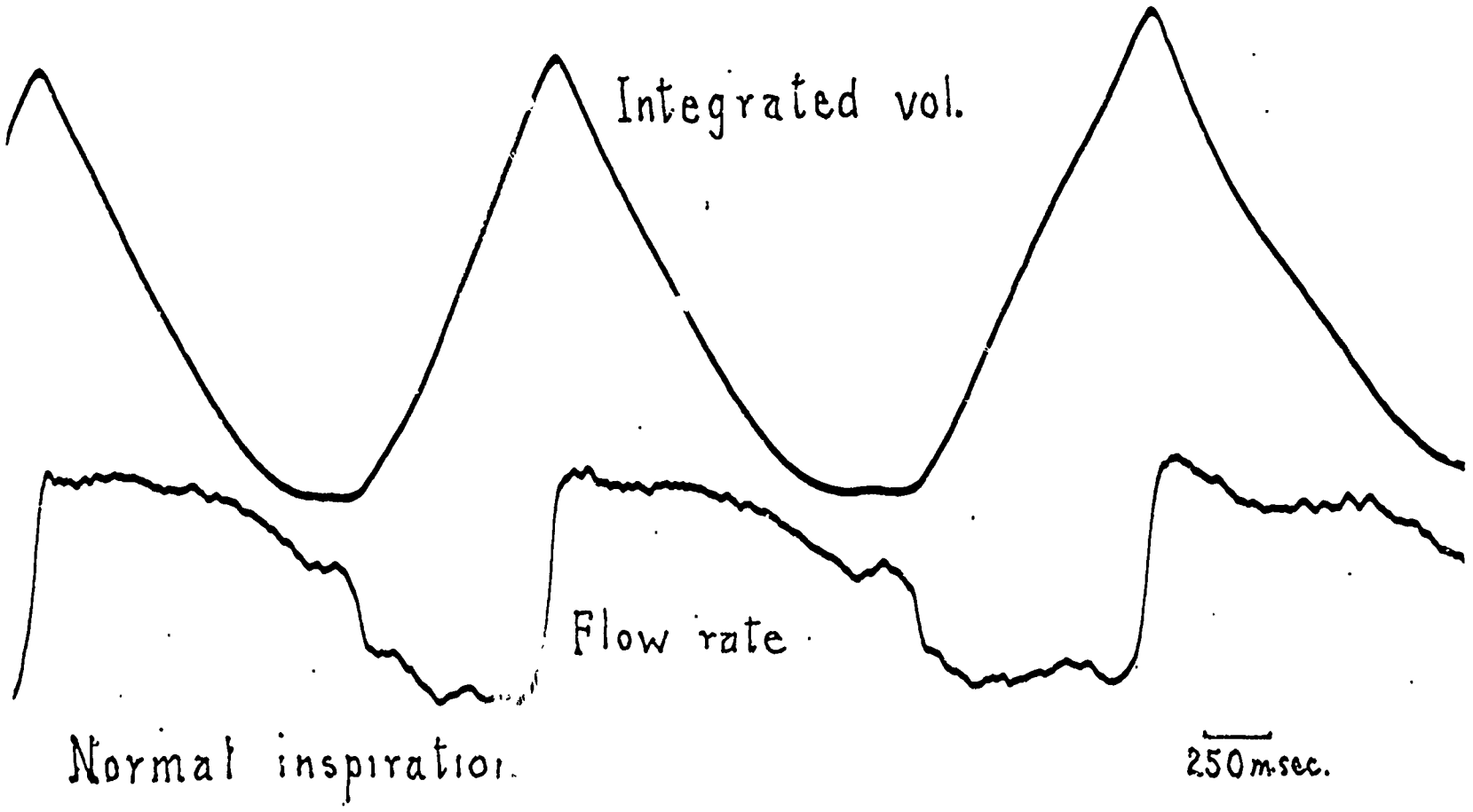


Fig. 12. Comparison of integrated vc [illegible] and flow ra[illegible] of air movements of dog (R) respiring room air between test stumu[illegible]. Downward deflections represent inhalation.

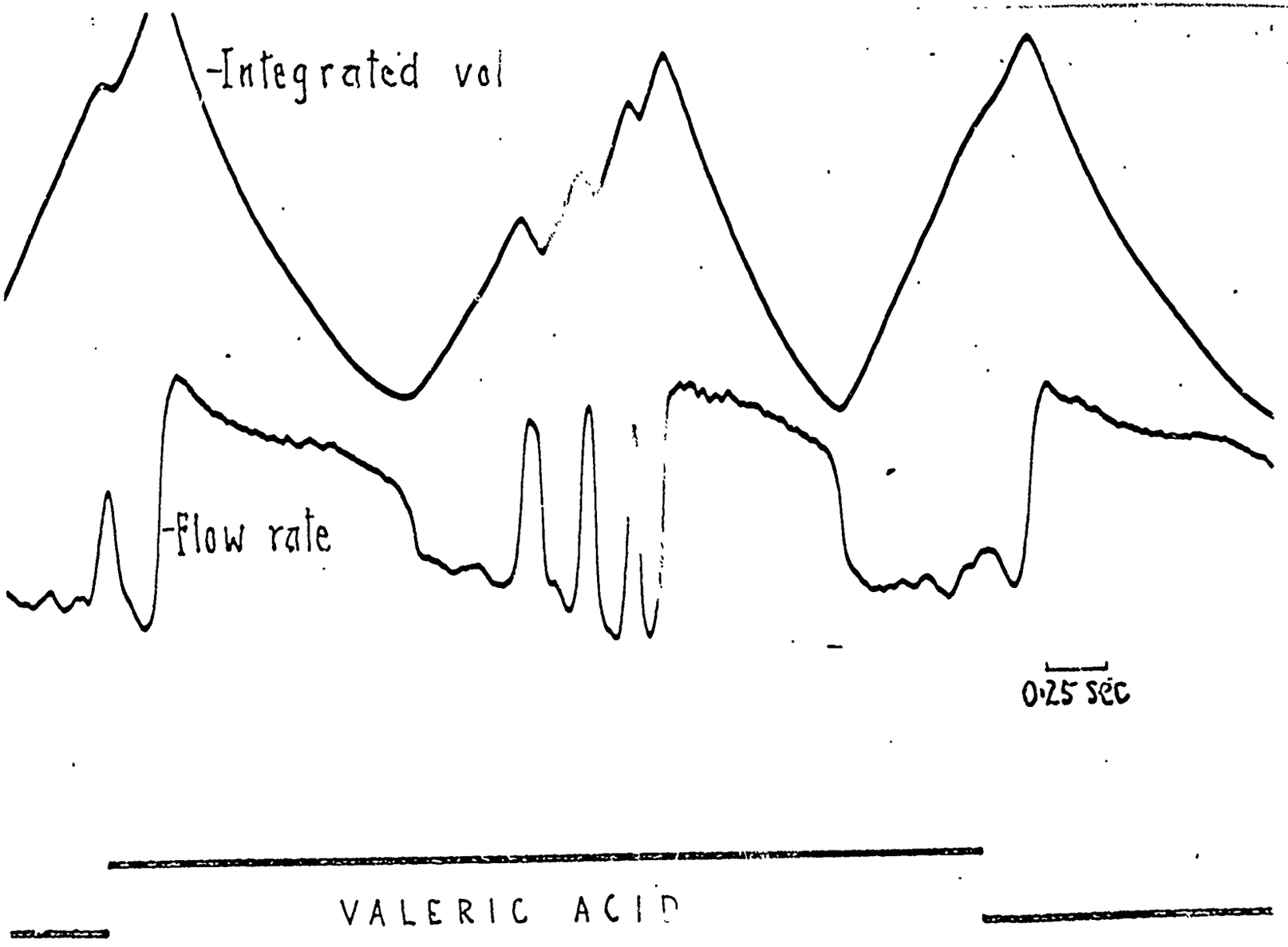
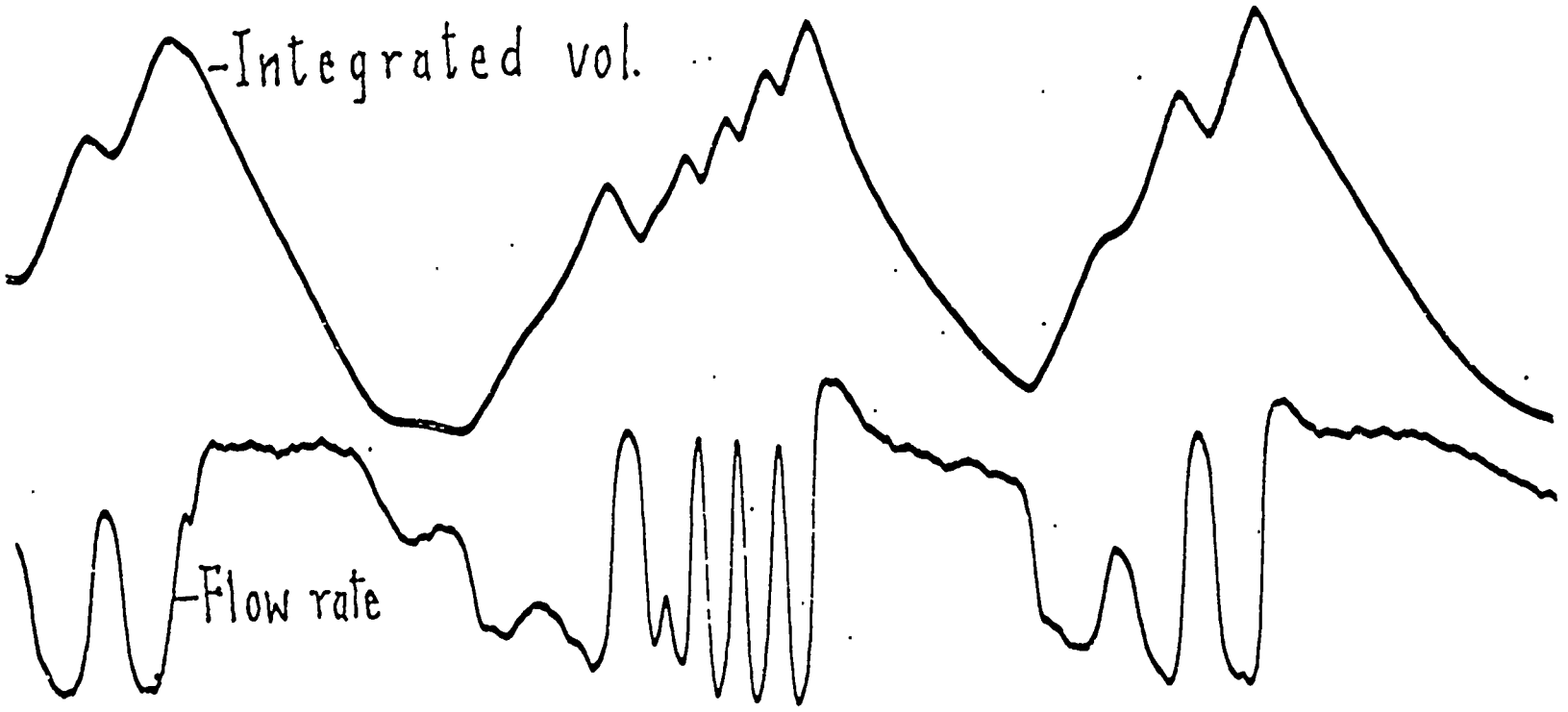
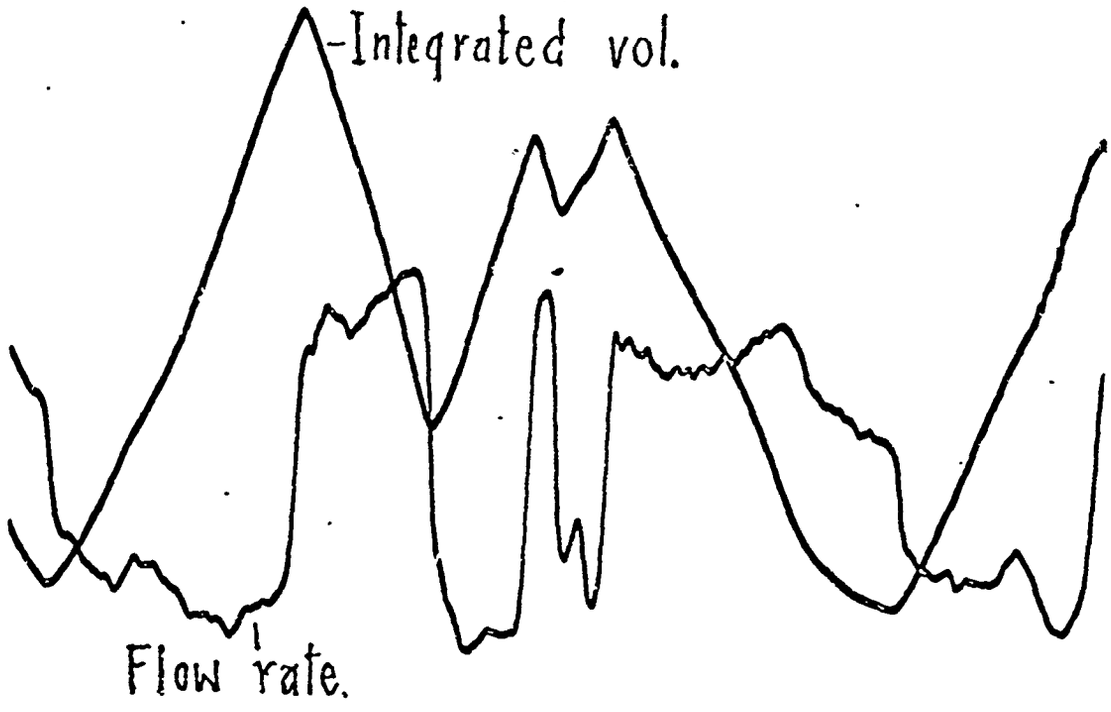


Fig. 13. Integrated sniff volume and flow rate of air movements of dog(R) sniffing valeric acid. Downward deflections of upper traces represent inhalation. Upward deflection of lower trace indicates stimulus delivery period.



CINEOLE

Fig. 14. Integrate sniff volume and flow rate of air movements of dog (R). Sniffingcireole Downward movements represent inhalation. (Same time scale as previous figures.) Upward deflection of lower trace indicates stimulus delivery period.



α IONONE

Fig. 15. Integrated sniff volume and flow rate of air movements of dog (R) sniffing α-ionone. Downward movements represent inhalation. (Same time scales as previous figures.)

2) Measurement of sniff parameters in dogs not performing a learned odor detection task

Introduction and methods

The data reported in the previous section was derived from dogs trained to detect odors. To provide some basis for comparison, however, it is important to know what differences occcur in the responses of dogs that are not actively engaged in a learned task. We therefore ran a further series of trials with dogs resting in a laboratory and breathing air at a relatively normal rate. Two of the three compounds tested were odors to which the animal had not previously been exposed. The compounds were α-ionone (which the dogs had been trained to detect); valeric acid and cineole.

Flow rate was measured with a Fleisch pneumotachograph attached by way of a specially-constructed aluminum cone (padded on the internal surfaces) and face mask. The dog had been habituated to wearing this device. The odors were held under the pneumotachograph in open neck bottles for approximately 1.5-2.0 seconds.

In addition direct measures of flow rate, "averaged" or "integrated" records of flow volume were obtained.

Results and Discussion

Examples of traces are shown in Figs. 12-15. In Figure 12 the dog is inspiring room air at a relatively normal rate (approximately one per second). Despite the erratic flow rate trace it is clear from the integrated readings that volume exchange is relatively constant and the breathing pattern stable.

In Figs. 13 and 14 an odor (to which the dog had not previously been exposed) was placed beneath the pneumotachometer intake. The preceding trace (not shown) indicated that until just before the marker indicating stimulus delivery was activated the animal was breathing normally. It is clear that the pattern of normal inspiration is interrupted in a very similar manner in both cases. However the integrated volume shows that the interruptions have little influence on the total volume exchanges (cf. Fig. 12). In contrast, the responses shown in Fig. 15 are to alpha-ionone - a compound to which this dog had been trained to respond in the three choice apparatus. In this case the interruption of the trace is more pronounced although it persists for less than one second (successive traces were normal).

These findings suggest that the casual sniffing of odors by the dog is a different phenomenon from the sniffing that occurs when dogs are actively seeking an odor source. They are more transient and may significantly influence volume exchange only when the odor is one to which the animal has been trained to respond or has otherwise acquired biological significance for the dog.

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Pietras, R. J. and Moulton, D. G. (1975) Influences of gonadal steroids on an odor detection task in the rat. In: Denton, D. (editor), Olfaction and Taste V. New York: Academic Press (in press).

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PRESENTATION

Moulton, D. G. (1974) Influences of gonadal steroids on an odor detection task in the rat. Paper given at the 5th International Symposium on Olfaction and Taste. Melbourne, Australia, October.