CYCLICAL VARIATIONS IN AVOIDANCE RETENTION:

EFFECT OF LIGHT CYCLE AND RELATIONSHIP TO FEAR

A THESIS

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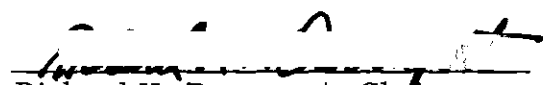
Studies

By

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CYCLICAL VARIATIONS IN AVOIDANCE RETENTION:

EFFECT OF LIGHT CYCLE AND RELATIONSHIP TO FEAR

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SUMMARY

It has been shown that the retention of a passive avoidance task in rats is cyclical if retention is assessed at successive six-hour intervals. To determine the possibility of entrainment to the alternating light-dark cycle of the housing environment, the present study investigated the effects of an alternating 12 hour light-12 hour dark cycle vs. constant illumination on retention. A pattern of high and low retention at successive six-hour intervals from 0 to 48 hours after training was found for both illumination conditions indicating that there was no entrainment to environmental light cycle. Sex and age differences were not significant. Furthermore, the relationship between fear and retention using differences in activity and defecation rates in training as compared to testing was assessed. Activity rates were lower in testing sessions as compared to training sessions when retention was high and remained the same when retention was low suggesting that fear was greater when retention was high. Defecation frequency was so low it was not a reliable index. The theoretical implications of cyclical variations in retention as well as the practical implications for animal learning research are discussed. Suggestions for future research are offered.

CHAPTER I

INTRODUCTION

Decay theories of memory have held that retention of a learned task can

be expressed as a negatively-accelerated monotonic function of time between

acquisition and performance. That is, retention is expected to decrease as time

from original learning increases. In recent years a number of experiments in

state-dependent learning have suggested that there are predictable variations in

retention over time and that these variations are more complicated than gradual

decay (Overton, 1964). One example of such a phenomenon is the "Kamin effect."

The Kamin Effect

Kamin (1957) found that the retention of an avoidance response varies

nonmonotonically as a function of time since original learning if a weak criterion

for learning was used. In his experiment, 25 avoidance training trials in a

shuttle-box were given to 6 groups of rats. Retention was assessed by testing

the subjects in the shuttle-box either immediately, .5, 1, 6, 24, or 456 hours

after training. The retention of original learning to subsequent relearning was

found to be a curvilinear function of the duration of the training-testing interval.

As indicated in Figure 1, retention declined from 0 to 1 hour and then rose from

1 hour to 19 days. This U-shaped function of retention has become known as

the "Kamin effect."

MEAN NUMBER OF AVOIDANCES

16

14

12

10

8

6

4

2

.5   1   6   24   456

RETENTION TIME (HOURS)
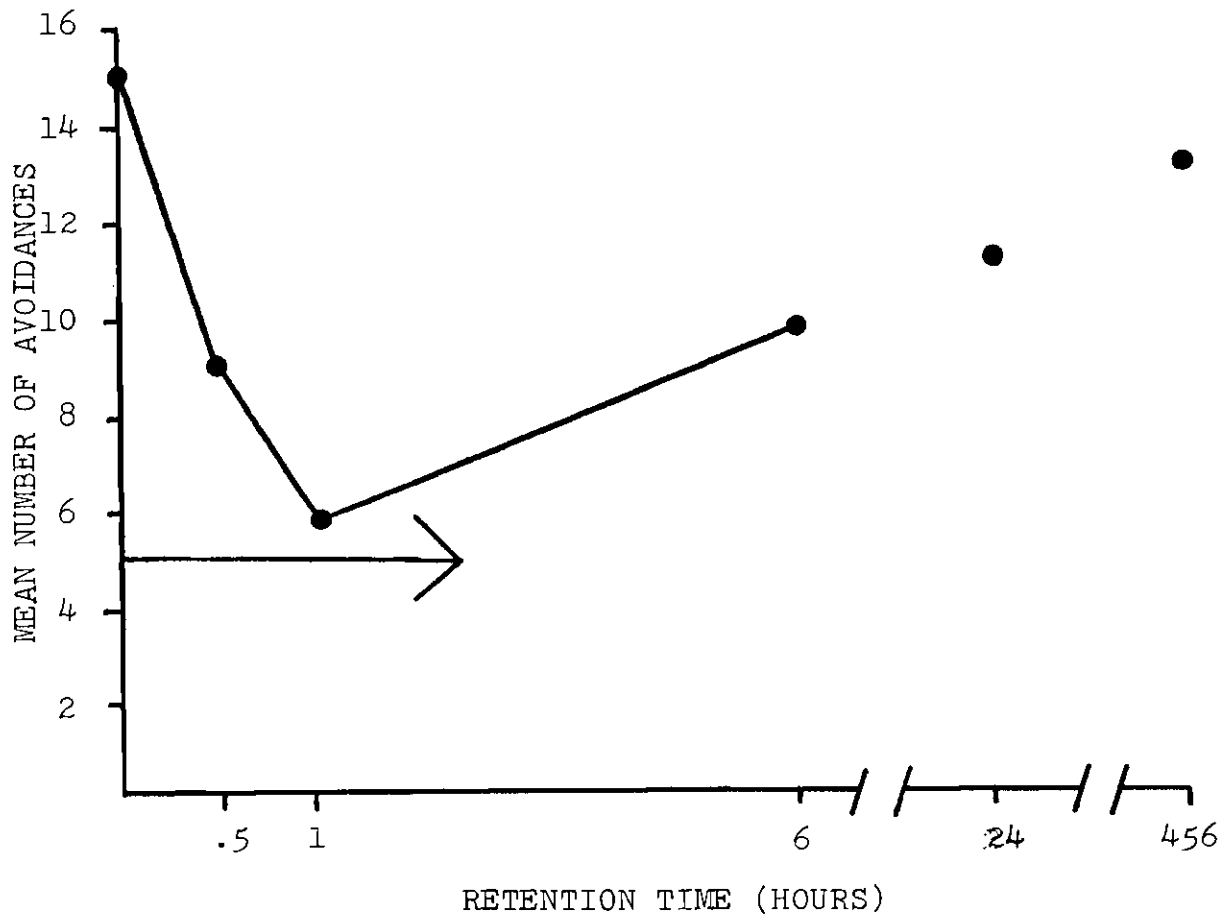
Figure 1. Adjusted Mean Number of Avoidances during Relearning as a Function of Retention Interval. (The arrow from the ordinate represents the grand mean number of avoidances during original learning. From L. J. Kamin. The retention of an incompletely learned avoidance response. Journal of Comparative and Physiological Psychology, 1957, 50, 457-460. Copyright (1957) by the American Psychological Association. Reprinted by permission.)

The Kamin effect has been replicated using both active and passive avoidance procedures with the time of the maximal retention deficit varying from 1 to 8 hours after training. These variations have seemed to result primarily from differences in procedure (active vs. passive avoidance tasks), apparatus, and a limited sampling of training-testing intervals.

Cyclical Variations in Avoidance Responding

More recently, Holloway and Wansley (1973a) found evidence that the nonmonotonic retention function characterizing the Kamin effect may be more complicated than originally believed by Kamin and possibly resulted from an incomplete sampling of training-testing intervals. Instead, an alternating pattern of high and low retention every 12 hours was found which suggested that "some biological factor (of unknown periodicity) may modulate fluctuations in the course of retention" (Holloway and Wansley, 1973a, p. 209). In their study, rats were randomly assigned to one of 13 conditions. Subjects in all conditions received passive avoidance training, but the conditions differed in the interval between the training and testing sessions, i.e., the 13 conditions were designated by their retention intervals which were 15 minutes, 6, 12, 18, 24, 30, 36, 42, 48, 54, 60, 66, and 72 hours. All groups were tested during the light phase of the light-dark cycle with approximately half of the subjects being tested early in the light cycle (10:00 A.M.) when activity level was assumed to be high while the remaining subjects were tested late in the light phase of the light dark cycle (4:00 P.M.) when activity level was assumed to be low. The two testing times

were chosen to determine if the activity cycle of the organism had an effect on avoidance performance.

The results of the Holloway and Wansley study are shown in Figure 2 and indicate that, on the average, more of the rats tested 15 minutes after training or at successive multiples of 12 hours after training (12, 24, 36, 48, 60, and 72 hours) retained the passive avoidance response than did rats tested 6 hours after training or at successive multiples of 12 hours from the 6 hour interval. Similar cyclical retention patterns have been found in active avoidance tasks (Holloway and Wansley, 1973b).

Holloway and Wansley suggested three possible explanations for the results. First, the absolute levels of activity might have influenced performance at testing directly. In other words, it was possible that retention was influenced by activity level such that good retention might be exhibited if the subject was tested when activity level was low and poor retention exhibited when activity level was high. This suggestion was unsupported, however, since there were no differences between subjects tested early and late in the light cycle when activity levels were assumed to be different and the same retention pattern was found for active and passive avoidance tasks.

Second, differences in the internal state of the organism at the time of testing from that of training might have contributed to the retention function. That is, physiological processes may provide altered organismic conditions or stimuli at the retention test which are different from those during original training. Support for this suggestion has been found by Stroebel (1967, as cited

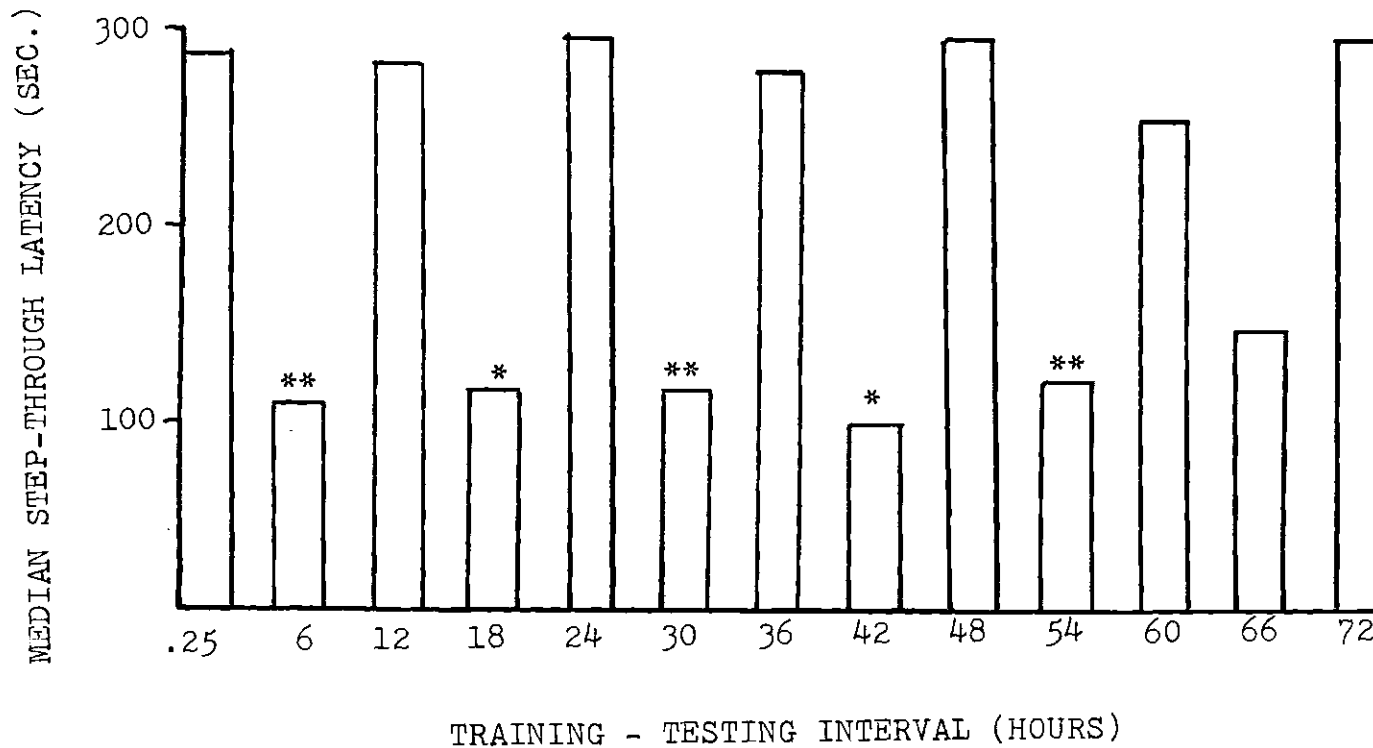Figure 2. Median Step-Through Latencies at Testing Sessions for each Training-Testing Interval. (Successive paired-group comparisons for each measure are *, P< .05; and **, P <.001. From F. A. Holloway and R. Wansley. Multiphasic retention deficits at periodic intervals after passive avoidance learning. Science, 1973, 180, 208-210, Fig. 1, 13 April 1973. Copyright (1973) by the American Association for the Advancement of Science.)

in Holloway and Wansley, 1973a) who reported that conditioned suppression of operant responses is stronger when tested at the same biological time (i.e., the time when biological processes are presumed most similar) as training than when tested at any other time of the 24 hour cycle. The critical stimuli have not been specified but might include modified central and peripheral nervous states of the organism, variations in hormonal functions, or cyclical environmental stimuli influencing the organism's own circadian rhythms.

Third, the shock may function as a Zeitgeber which entrains some cyclical physiological process which influences retention. That is, something analogous to a stress response or some other psycho-physiological reaction induced by the training procedure may, itself, become associated with a rhythmic variable resulting in cyclical avoidance performance.

It was suggested earlier that the nonmonotonic retention function demonstrated by Kamin (1957) and Holloway and Wansley (1973a, 1973b) may be examples of the same phenomenon with the single retention deficit characterizing the Kamin effect resulting from a limited sampling of training-testing intervals. If such is the case, then the nonmonotonic retention patterns of Kamin and Holloway and Wansley may have a common underlying mechanism. A number of hypotheses have been proposed as possible explanations for the Kamin effect which may be applicable to the Holloway and Wansley findings. These theoretical approaches are reviewed in the following sections of this paper. Critical analyses concerning their generality and applicability in accounting for cyclical retention functions are reserved for the discussion section of this paper.

The Multi-Storage Theory and Nonmonotonic Retention

Halstead and Rucker (1968, 1972) have offered an explanation of the Kamin effect based on a three stage model. According to this model, memory consists of: (1) a dynamic or labile short-term stage; (2) an intermediate stage during which time the dynamic trace is dissipating and a more permanent trace is developing; and (3) a permanent or consolidated long-term stage. These stages are hypothesized to operate sequentially with independent growth and decay rates such that as the trace in one stage is dissipating, the trace would be developing in the next stage. Performance at any time was believed to reflect the sum total of retention in all of the stages. Recall would be exhibited when this sum total was above a hypothetical threshold level. If the memory trace is interfered with prior to its development in the consolidated stage, say be electroconvulsive shock, retention of the response would be reduced.

Halstead and Rucker have attributed the poor retention characterizing the Kamin effect to occur when the intermediate stage of memory was dissipating and consequently at a low, sub-threshold level, and prior to the development of the trace in the more permanent, consolidated stage. That is, poor retention would be exhibited when the sum of retention in the intermediate and consolidated stages fall below the threshold level necessary for recall. Presumably this is due to the unequal rates of decay and growth of the traces in these two stages. At shorter training-testing intervals, the trace is above threshold level in the intermediate stage. As the training-testing interval increases beyond the time of the retention deficit, more information is entering the consolidated stage and

retention of the avoidance response would be exhibited when the developing trace in the consolidated stage passes the hypothetical threshold level.

The State-Dependent Theory and Nonmonotonic Retention

Klein and Spear (1969, 1970) and Spear, Klein, and Riley (1971) have suggested a retrieval failure explanation for the Kamin effect. According to this interpretation, retention is dependent on the presence of organismic retrieval cues during the retention test that are the same or similar to the attributes of original learning. The poor retention was attributed to the reduced availability of retrieval cues which resulted from shock-induced physiological changes at intermediate retention intervals. It was hypothesized that these physiological changes had not yet occurred at shorter retention intervals and had dissipated at longer intervals. Their basic argument was that poor avoidance responding at intermediate intervals resulted from the presence of novel, organismic stimuli that were not previously associated with the avoidance response during training. These altered organismic stimuli produced retrieval failure. It should be noted that a similar explanation based on altered organismic stimuli has been proposed by Overton (1964) to account for the dissociation phenomenon in drug state-dependent learning.

While the altered organismic stimuli were not stated, other investigators have implicated the neuroendocrine system and parasympathetic nervous system as the possible physiological mechanisms mediating the Kamin effect. These theories are presented in the following two sections.

Adrenal Corticosteroids and Nonmonotonic Retention

Brush and Levine (1966) have suggested that the functioning of the neuro-
endocrine system may be the physiological basis for the Kamin effect. Accord-
ing to these investigators, the poor retention found at about 1 hour was directly
related to corticosteroid levels in response to the shock-induced stress of
training. It was found that the decrease in shuttle-box retention was correlated
in time with a corresponding decrease in the plasma concentration of corticos-
terone. Good retention, in turn, was correlated with the return of corticosterone
levels to those levels present during training. The retention deficit was abol-
ished when high corticosteroid levels were maintained by either injections of
adrenocorticotropic hormone (ACTH) or hydrocortisone replacement after train-
ing (Levine and Brush, 1967). From these results, it was hypothesized that the
retention deficit characterizing the Kamin effect resulted from variations in
corticosterone levels which provided altered organismic cues at the time of
testing from those at training.

Parasympathetic Overreaction and Nonmonotonic Retention

An alternative physiological explanation for the Kamin effect has been
based on Brush, Myer, and Palmer's (1963) suggestion that the Kamin effect
results from a parasympathetic overreaction to the stress of original training
and had little to do with retention, per se. That is, the shock-induced stress
of avoidance training is detected by the brain which triggers a release of epineph-
rine peripherally. The brain detects this release of epinephrine via a neuronal

feedback system from altered peripheral sites. This detection process involves

a cholinergically-based link in the brain which results in response suppression

(Manto, 1967; Carlton, 1969). It follows that the Kamin effect could result from

this parasympathetic rebound reaching a maximum at about one hour and then

dissipating as time increases.

Support for this interpretation has been provided by Thompson and

Neilson (1972). They investigated the differential effects of injections of scopola-

mine hydrobromide (a drug which reduces the muscarinic, parasympathetic

actions of acetylcholine in the peripheral nervous system and the brain, i.e., a

cholinergic inhibitor) and methylscopolamine (a drug with the same peripheral

effects as scopolamine but does not cross the blood-brain-barrier and, conse-

quently, has little, if any, effect on the brain) on avoidance performance as a

function of time since training. It was found that the Kamin effect was exhibited

by rats treated with methyl scopolamine and was not exhibited by rats treated

with scopolamine hydrobromide. Since scopolamine hydrobromide crossed the

blood-brain-barrier and presumably acted as a cholinergic-inhibitor, it was

proposed that central cholinergic activity, producing response suppression, was

a possible cause for the Kamin effect.

The Incubation of Anxiety Theory and Nonmonotonic Retention

An alternative explanation for the Kamin effect has been proposed by a

number of investigators based on the incubation of anxiety theory originally

formulated by Bindra and Cameron (1953). Three different incubation theories

for the Kamin effect have been suggested. Kamin (1957) interpreted his results

in terms of two independent processes. The decline in retention during the

first hour was attributed to a "forgetting" process which reaches asymptote by

one hour. It was suggested that a large part of this forgetting might have been

due to a disruption of set and postural adjustment resulting from the removal of

the subject from the training situation. The increase in retention after one hour

was attributed to an incubation of fear, i. e., the fear associated with the avoid-

ance situation increased with the time that had elapsed between training and the

first re-exposure to that situation. This increase in fear was believed to have

resulted in improved avoidance behavior. No evidence, however, was provided

in support of such an interpretation.

Denny (1958) and Denny and Ditchman (1962) have provided an alternative

explanation based on the incubation of anxiety theory and Denny's (1958) obser-

vation that animals tested one hour after training typically freeze. This freezing

behavior is incompatible with the active avoidance response. They then suggested

that after a delay of one hour, the anxiety dissipates and retention of the avoidance

response is present after longer retention intervals. That is, increases in anxi-

ety induced by the shock during training leads to general response suppression

which is maximal during intermediate retention intervals and interferes with

avoidance behavior.

While anxiety or fear is a common factor intrinsic to both Kamin (1957)

and Denny (1958), the actual application of the anxiety notion is quite different

in each. According to the former view, an incubation of fear results in improved

avoidance behavior at intervals greater than one hour. The latter view, however, attributes the poor avoidance behavior at one hour to the incubation of fear which results in response suppression. Denny's (1958) hypothesis was questioned by Pinel and Cooper (1966) who have suggested an alternative explanation based on anxiety. Pinel and Cooper believed that the decline in avoidance performance early in the Kamin effect was related to a decrease in fear rather than an increase in fear as suggested by Denny (1958). Furthermore, Pinel and Cooper have attributed the improved avoidance behavior after about one hour to an increase in fear, the same as proposed by Kamin (1957).

In review of the anxiety theories concerning the nonmonotonic retention effect, the initial decline in retention has been attributed to: (1) forgetting (Kamin, 1957); (2) an incubation of fear (Denny, 1958; Denny and Ditchman, 1962); and (3) a decrease in fear (Pinel and Cooper, 1966). The increase in retention has been attributed to: (1) an incubation of fear (Kamin, 1957; Pinel and Cooper, 1966) and (2) a decrease in fear (Denny, 1958; Denny and Ditchman, 1962). Thus, if an anxiety notion is to be useful as an explanation of nonmonotonic retention, it would seem worthwhile to investigate variables correlated with fear and determine the relationship of fear to performance at various retention times.

The Present Study

The general aim of the proposed experiment was to further investigate the nonmonotonic retention pattern and to study associated variables. Specifically, the present study was designed to: (1) replicate the Holloway and Wansley (1973a) experiment demonstrating cyclical retention; (2) determine the effect of two

conditions of light cycle on the phenomenon; and (3) assess the relationship between fear and retention.

The procedure used in the present study was similar to that used by Holloway and Wansley (1973a). Rats were assigned to experimental conditions defined by the interval of time between the performance of an avoidance response and the subsequent retention of that response. The first major difference in the proposed study from the Holloway and Wansley design was in the type of environmental light cycle used in housing the animals. In the Holloway and Wansley study, subjects were housed in an alternating 12-hour light - 12-hour dark environment. It was possible that the cyclical pattern of retention found by Holloway and Wansley resulted from the cyclical nature of the illumination of the housing environment such that the alternating light-dark cycle may have caused retention to occur in rhythmic patterns.

Furthermore, in their study, testing occurred at one of two times during the light phase of the light-dark cycle to examine the role of activity level on avoidance. The use of the two testing times, however, may have been confounded with the alternating light-dark cycle such that all subjects in the 15 minute, 24, 48, and 72 hour retention interval conditions were trained and tested during the light phase of the light-dark cycle. These subjects showed good retention. Subjects trained in the 12, 36, and 60 hour conditions were trained only during the dark phase of the light-dark cycle. In training, the subject was placed in an illuminated test chamber which might have reset their internal mechanisms controlling rhythmic activities (their biological clocks). These subjects also

exhibited good retention. Subjects trained in the 6, 18, 30, 42, 54, and 66 hour retention interval conditions had approximately an equal number of subjects trained during either the light or dark phase of the light-dark cycle. It was interesting that the number of subjects in these retention interval conditions reaching criterion on the retention test was low compared to the other conditions. Holloway and Wansley did not report whether the phase of the light-dark cycle during which training occurred influenced retention directly, thus necessitating an examination of the effect of the light-dark cycle during training on subsequent retention.

To examine the role of light cycle on cyclical avoidance performance, two groups of rats were used in the present study. The first group was housed in an environment with an alternating light-dark cycle as used by Holloway and Wansley. The primary purpose of this group was to replicate the original Holloway and Wansley experiment of cyclical retention. A second group was trained and tested on an identical task with the exception that they were housed in continuous illumination which enabled both training and testing to occur during the same phase of the light-dark cycle (i.e., the light phase) for all retention intervals and precluded the possibility of alternating light-dark phases from influencing retention.

In the present study, two behaviors correlated with fear were also studied. The response measures taken at all training and testing sessions were: (1) a defecation score based on the number of fecal boluses and (2) an activity score measured by the animal's ambulation. Parker (1939, as cited in Hall, 1941)

has found that defecation is a fairly general response to a wide variety of "fear-inducing" stimuli such as a loud sound, sudden dropping or other loss of support, tilting and sliding, forced swimming, and immobilization. The correlations of defecation frequency on all of these tests were high and positive ranging from .60 to .90. It has also been reported that exploratory behavior, as inferred from the activity score, and fear are negatively correlated (Hayes, 1960). While ambulation and defecation are not direct measures of fear, they are correlated with a psychophysiological state that is believed to be fear-induced and are used as measures to infer fear. It was possible that defecation and ambulation would reflect the existence and quantify the degree of fear of the subjects during training and testing. These measures should provide a firmer base for testing the relation between fear and nonmonotonic retention than the casual observation of freezing (Denny, 1958) or speculation (Kamin, 1957).

In summary, the proposed experiment was designed to examine the cyclical retention of an avoidance task under two lighting conditions, an alternating 12 hour light-dark cycle and a constant light environment. In addition, ambulation and defecation measures were taken to enable the assessment of fear at the various retention intervals in order to test the anxiety theory for the Kamin effect. The subjects were rats. The dependent variables were: (1) avoidance performance; (2) ambulation; and (3) defecation. The independent variables were: (1) the two conditions of illumination and (2) the interval between initial avoidance training and the test for retention.

Statement of the Hypotheses

I. The first experimental hypothesis is that the cyclical retention pattern found by Holloway and Wansley (1973a) was an artifact related to differences in the phase of the light-dark cycle present during training and that of testing. To examine this possibility, the performance of subjects trained during the light phase of the cycle were compared with those trained during the dark phase.

II. The second hypothesis is that the cyclical retention pattern resulted from the alternating illumination conditions of the housing environment. This possibility was investigated by examining the effect of an alternating light-dark cycle vs. constant light on avoidance performance.

III. The third hypothesis tested concerned the relation between fear and retention. It was hypothesized that high fear as measured by ambulation and defecation would occur in test conditions where there was high retention as compared to low retention conditions.

CHAPTER II

METHOD

Subjects

Ninety male and female Sprague-Dawley albino rats 60 to 150 days of age

(mean age = 111.5) from the Georgia Tech colony served as subjects. They

were randomly divided into two groups of 45 rats each. One group was housed,

2 rats per cage, in a 12 hour light - 12 hour dark condition while the second was

housed, 2 rats per cage, in a constantly illuminated environment. Both groups

lived for a two week pre-experimental adaptation period to accustom them to

their respective environments which were continued throughout the experiment.

This adaptation procedure was used by Holloway and Wansley (1973a). The 45

subjects in each group were assigned to one of the nine experimental conditions

so that there were 5 rats from each group in each of the conditions with at least

2 rats of each sex per condition. Litter mates were distributed across conditions.

All subjects were given free access to food (Purina rat chow) and water in their

housing areas.

Housing and Testing Environments

The light-dark environment was characterized by an alternating 12 hour

light period (8:00 A.M. to 8:00 P.M.) followed by a 12 hour dark period (8:00 P.M.

to 8:00 A.M.). The average illumination of the cage area was 30 foot-candles

during the light period and the average temperature was 72° F. The length of each cage was 35 cm. and the width was 23 cm. The constant light environment had an average illumination of 28.5 foot-candles and an average temperature of 74° F at the cage area. The cages in this area had a length of 24 cm. and a width of 20 cm. The cages were cleaned once a week at the same time (4:00 P.M.). Feeding and watering occurred daily, also at the same time (4:00 P.M.). The test room had an average temperature of 74° F.

Apparatus

The avoidance apparatus consisted of two chambers. The open field start chamber was circular with a radius of 62 cm. providing a total area of approximately 1.2 sq. m. Lines were painted on the floor of this chamber forming a grid of 49 squares which were used in determining the ambulation score. Each of the squares of the grid had an area of 245 sq. cm. The start chamber was illuminated by overhead flourescent lights which provided even illumination (80 foot-candles). At one end of the start chamber was an opening (measuring 9 cm. in height and 10.5 cm. in width) into a darkened, rectangular shock chamber measuring 20 cm. in length, 15 cm. in width, and 21 cm. in height. All walls of the shock chamber were opaque and the floor was construc-ted of 18 parallel metal bars 15 cm. long spaced 1.1 cm. apart (Ralph Gerbrands Co., Model 0). These bars when activated delivered a .5 ma electric shock produced by a Grason-Stadler shock generator (Model E1064GS). All times were recorded by electronic timers. All timers and the shock generator were controlled by hand.

Procedure

All subjects were given a one trial shock avoidance task in a room sepa-

rate from the housing areas. Training occurred either .25, 6, 12, 18, 24, 30,

36, 42, or 48 hours prior to testing, depending on the experimental condition to

which each animal was assigned. Testing occurred at 4:00 P.M. for all subjects.

On the training trial, the subject was placed in the center of the start

chamber facing away from the shock chamber. A timer was activated at this

time which was stopped when the subject completely entered the shock chamber.

The time between placing the subject in the start chamber and its entering the

shock chamber, the step-through latency, was recorded. While in the start

chamber, the number of lines crossed by the rat's nose and number of fecal

boluses for each successive 10-second interval were counted. As soon as the

rat completely entered the shock chamber (including its tail), the shock was

administered and continued until the rat re-entered the start chamber when it

was immediately removed by the experimenter and returned to its home cage

for a period of time determined by its experimental condition. Although it had

been planned that subjects not entering the shock chamber within 15 minutes

after placement in the start chamber would be discarded as nonresponders,

none of the subjects took longer than 160 seconds.

The procedure for retention testing was similar to that used for training

except for the shock. Ambulation, defecation, and step-through latencies were

recorded as during training. Subjects not entering the shock chamber within

300 seconds (as used by Holloway and Wansley, 1973a) or 10 times their own

step-through latency during training (whichever was longer) were scored as

retaining the avoidance task.

CHAPTER III

RESULTS

The three variables measured during training and testing sessions for all

subjects included: (1) the step-through latency; (2) the ambulation rate (number

of lines crossed per unit time); and (3) the defecation rate (number of fecal boluses

per unit time). The .05 probability level was used as the indication of statistical

significance.

To determine whether any differences existed on the training step-through

latencies across groups and conditions resulting from differences in training times

or housing environments, a simple one-way analysis of variance was performed

(Kirk, 1968). No significant differences were found $(F(8, 81) = 1.23)$.

For the purpose of analyzing the testing step-through latencies, the

behavioral criterion (10 times the training step-through latency) was discarded in

favor of the time criterion (300 seconds). This was done to eliminate extremely

long latencies from data analysis and was judged permissible in that only one of

the 90 subjects had a testing step-through latency greater than 300 seconds and

failed to reach the behavioral criterion. The number of subjects reaching the 300

second criterion for each housing illumination group as a function of retention

interval are shown in Figure 3. The median testing step-through latencies for

each housing illumination group as a function of retention interval are shown in

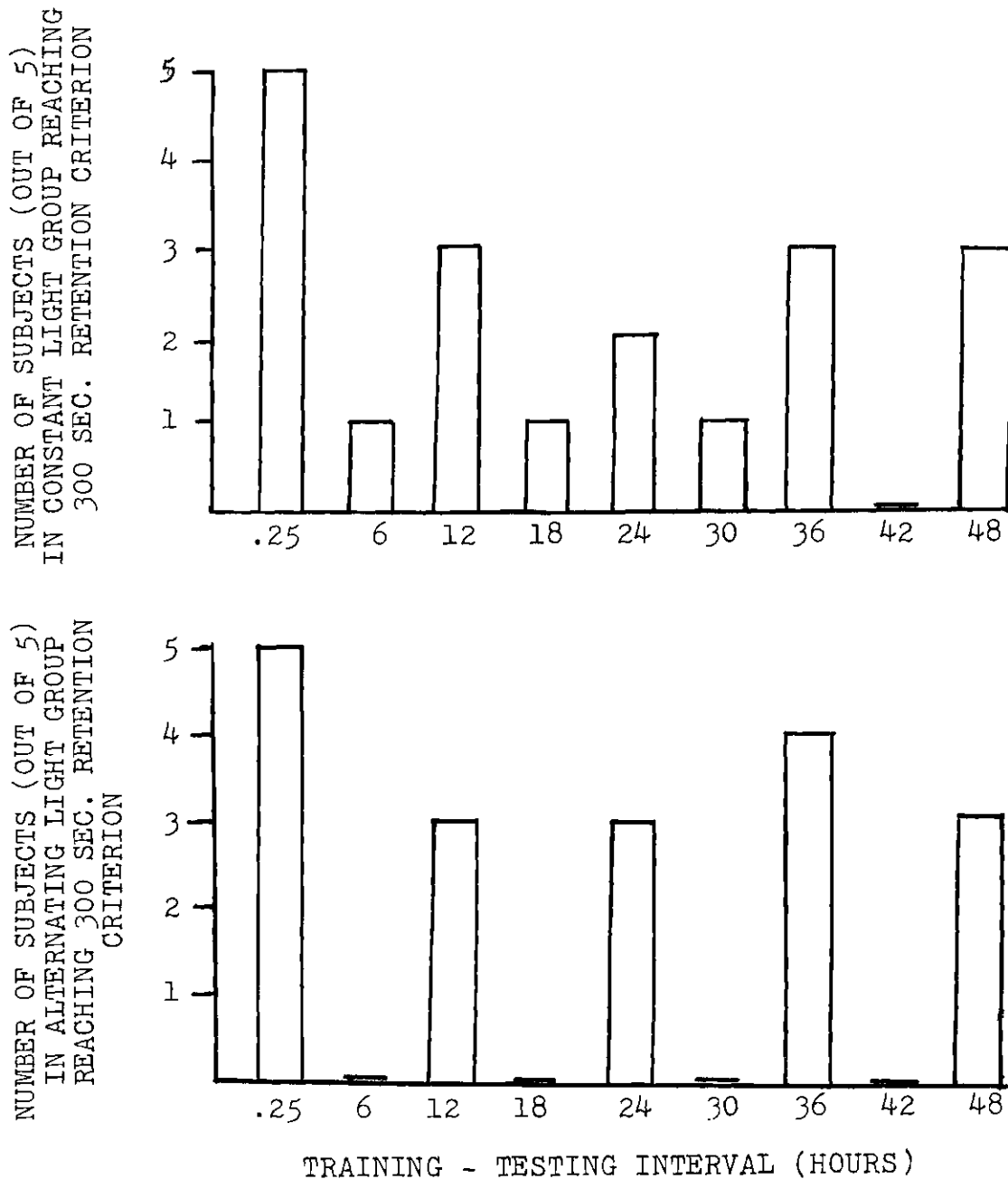Figure 4. It is clear that differences existed for both housing illumination groups

Figure 3. Number of Subjects Reaching 300 Second Criterion of Retention for Each Housing Illumination Group as a Function of Retention Interval.
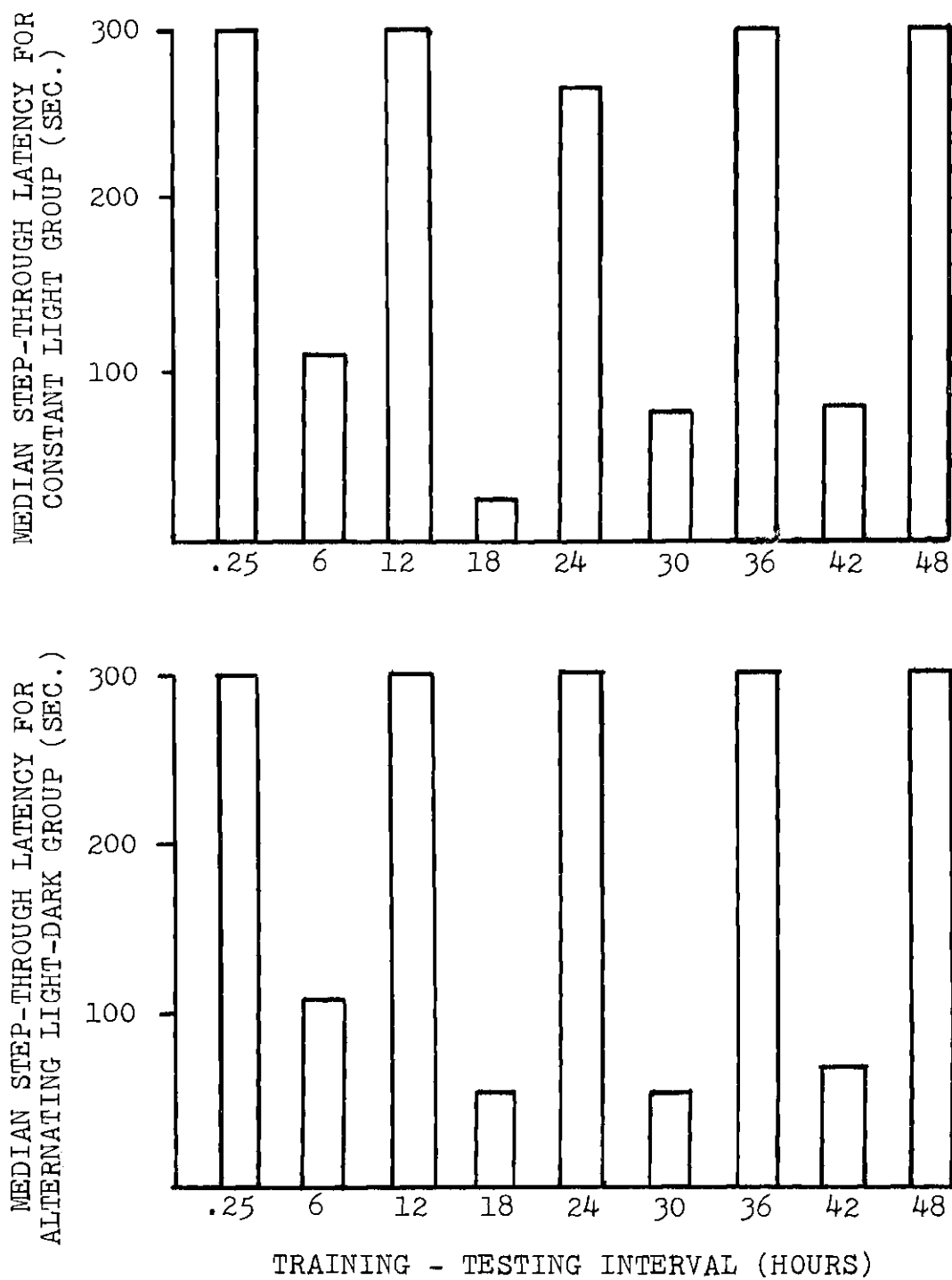
Figure 4. Median Step-Through Latencies for Each Housing Illumination Froup as a function of Retention Interval.

in the number of subjects reaching criterion and in the median step-through

latencies with greater numbers of subjects reaching criterion and longer medain

step-through latencies for the .25, 12, 24, 36, and 48 hour retention interval

conditions as compared to the 6, 18, 30, and 42 hour conditions. These retention

interval conditions will be subsequently referred to as full cycle and half cycle

conditions, respectively.

To determine whether significant differences existed between the two

housing illumination groups on the median testing step-through latencies, a

Friedman rank sums two-way analysis with a correction for tied scores was per-

formed (Hollander and Wolfe, 1973). No significant differences were found between

the group housed in the alternating light-dark environment and the constant illumi-

nation environment $(S(1)=.2)$. That no differences existed between the housing

illumination groups permitted the pooling of data from both groups for subsequent

analyses.

To determine whether there were significant differences among the nine

retention interval conditions on the pooled step-through latencies, a Kruskal-

Wallace one-way analysis with a correction for tied scores was performed

(Hollander and Wolfe, 1973). The analysis showed that significant differences did

exist $(H(8)=35.07, p < .05)$. To determine where such differences existed, two

separate Kruskal-Wallace analyses with corrections for tied scores were then

performed. The first analysis was on the testing step-through latencies of the

full cycle retention interval conditions. The analysis revealed no differences

among these retention intervals $(H(4)=5.14)$. The second Kruskal-Wallace analysis

was performed on the half cycle retention interval conditions. This analysis also indicated that there were no differences among these retention intervals (H(3)=3.06). Because differences did exist on the Kruskal-Wallace analysis of all nine retention intervals and no differences existed within either the full cycle or half cycle intervals, it can be concluded that the differences found in the initial analysis resulted from differences between the full cycle and half cycle retention interval conditions. These results indicate that retention as measured by the testing step-through latencies was significantly better at the full cycle retention intervals than at the half cycle intervals for both housing illumination groups and that no differences existed between the illumination conditions.

To determine the effect of the phase of the light-dark cycle during training on subsequent performance for the group of subjects housed in the alternating light-dark environment, two two-tailed Mann-Whitney U-tests for differences between independent samples were used (Bruning and Kintz, 1968). The first analysis compared the testing step-through latencies for subjects trained in the dark with those trained in the light for the half cycle retention interval conditions. No differences were found between the two training phases of the light-dark cycle (U(10,10)=48). A similar analysis performed on the full cycle testing step-through latencies also failed to reveal differences between training in the light phase and training in the dark phase (U(10,10)=66). Thus, the phase of the light-dark cycle during training had no significant effect on subsequent retention.

To determine whether there were concomitant variations in ambulation rates with retention, difference scores between ambulation rates obtained during

testing and training were calculated for each subject. The pooled data for each

retention interval condition were then analyzed separately using t-tests for dif-

ferences between a sample mean and a population mean (assumed to be zero,

Hayes, 1973). A summary table of the obtained t-tests for each of the groups is

shown in Table I. All full cycle conditions had a significant decrease in ambula-

tion rates on the testing session as compared to the training session while only

one of the half cycle conditions (the 18 hour retention interval condition) differed

significantly from zero.

While it was also intended to analyze the defecation data in a manner

similar to the ambulation data, so few subjects defecated on either training or

testing sessions that the analysis of such data was judged not to be fruitful.

Male and female rats were used in this study. It was possible that sex

differences might exist in the experiment. Two, two-tailed Mann-Whitney U-tests

were used to determine if sex differences existed on the testing step-through

latencies. The first analysis was performed on the full cycle conditions. No sex

differences were found (U(25, 25)=291.5). The second analysis performed on the

half cycle conditions also revealed no sex differences (U(20, 20)=151).

Age of the subjects ranged from 60 to 150 days. It was possible that age

might be related to retention. To examine this possibility, the subjects were

divided into six age ranges based on their age at training. Each range constituted

a 15 day interval. Two Kruskal-Wallace analyses with a correction for tied scores

were then performed on the testing step-through latencies across the age ranges.

The first analysis was performed on the full cycle conditions and yielded no

Table I. Summary Table of the t-Tests for Differences Between

Sample and Population Mean as a Function of Retention Interval

Retention Interval	t	d.f.	p
.25	50.97*	9	< .05
6	1.61**	9	> .05
12	4.92*	9	< .05
18	2.49**	9	< .05
24	39.87*	9	< .05
30	1.93**	9	> .05
36	10.17*	9	< .05
42	1.61**	9	> .05
48	3.05*	9	< .05

* One-tailed analysis, critical t(9) = 1.83.

** Two-tailed analysis, critical t(9) = 2.26.

significant differences between age categories (H(4)=3.40). A similar analysis performed on the half cycle conditions also revealed no significant age differences (H(3)=2.89).

In summary, the results indicate that there are cyclical variations in retention as shown by the number of subjects reaching criterion and the step-through latencies. It was also found that there are cyclical variations in ambulation which are associated with retention.

CHAPTER IV

DISCUSSION

The results of the present study confirm and extend the findings of

Holloway and Wansley (1973a), i.e., a pattern of alternating high and low reten-

tion of a task at successive six hour intervals after original training on that task.

The robustness of this cyclical retention phenomenon was emphasized by showing

that it occurred in rats: (1) housed in an alternating light-dark environment;

(2) housed in continuous illumination which reduced the circadian effects of an

alternating light-dark cycle; (3) of both sexes; and (4) of ages from 60 to 150 days.

Ambulation and defecation, two measures which in other studies have

been shown to be correlated with the psychophysiological state of fear, were

examined in the present study to determine if there was a concomitant variation

of fear and retention. The results suggest that fear, as measured by ambulation

rates, was greater when avoidance retention was high at full cycle conditions than

when retention was low at half cycle conditions. Defecation was so infrequent

that it was not a reliable index.

Cyclical variations in retention may be of theoretical importance. One

of the purposes of the present study was to determine the appropriateness of the

incubation of anxiety theory in accounting for cyclical retention. According to the

various interpretations of this theory, low retention is attributed to: (1) an incu-

bation of fear (Denny, 1958; Denny and Ditchman, 1962) or (2) a decrease in fear

(Pinel and Cooper, 1966). High retention has been attributed to: (1) an incubation of fear (Pinel and Cooper, 1966; Kamin, 1957) or (2) a decrease in fear (Denny, 1958; Denny and Ditchman, 1962). Since the present study suggests that fear was greater when retention was high at full cycle intervals than when retention was low at half cycle intervals, the results were not inconsistent with the Pinel and Cooper hypothesis. It should be noted, however, that the ambulation measure on which this suggestion was made could have resulted from influences other than fear.

The multi-storage theory of memory proposed by Halstead and Rucker (1968; 1972) does not adequately account for the cyclical retention function. Their theory predicts that poor retention would occur at only two periods after initial training: (1) the time when the dynamic trace is dissipating and the intermediate trace is developing and (2) the time when the intermediate trace is dissipating and the consolidated trace is developing. Since the consolidated stage is presumed permanent, repetitive cyclical retention deficits would not be predicted. Thus, the present results indicate that the multi-storage theory is untenable in accounting for periodicities in retention.

Two theories have been proposed to account for variations in retention based on the state-dependent notion that retention depends on the similarity of organismic cues in the training and retention test situations. The retention deficit of the Kamin function is attributed to the reduced similarity of these cues at intermediate retention intervals. Both the parasympathetic overreaction theory and the adrenocortical theory suggest that shock-induced physiological changes occur which result in altered organismic cues. The parasympathetic overreaction

theory proposes that shock causes increases in epinephrine which results in a parasympathetic rebound, the consequence of which is reduced behavioral activity (Brush, Myer, and Palmer, 1963; Manto, 1967; Carlton, 1969; Thompson and Nielson, 1972). The adrenocortical theory suggests that shock causes a decrease in corticosterone (thus, an increase in ACTH) which results in a state-dependent retention failure (Brush and Levine, 1966; Levine and Brush, 1967).

For either of these theories to adequately explain the cyclical retention pattern, it must be demonstrated that either epinephrine, corticosterone, or ACTH levels have a periodicity of approximately 12 hours. Corticosterone (hydrocortisone in humans) and ACTH have been found to have a circadian rhythm in a number of species including man (Halberg, Halberg, and Haus, 1974; Leach and Campbell, 1974; Lakatua, Haus, Gold, and Halberg, 1974). Cyclicities in epinephrine levels, however, have not been found.

It should be noted that these theories do not adequately account for some of the experimental findings of variations in retention. The parasympathetic overreaction theory is a "performance" theory based on shock-induced response suppression and is unable to parsimoniously account for high retention on active as well as passive avoidance tasks at full cycle intervals (Holloway and Wansley, 1973a; 1973b). The adrenocortical theory is unable to explain variations in retention in adrenalectomized rats (Snider, Marquis, Black, and Suboski, 1971) and in hormonally underdeveloped rats (Klein and Spear, 1969). Furthermore, the adrenocortical theory predicts that poor retention should occur when ACTH levels are high. Levine (1971) has found that fear is characterized by high ACTH levels.

If the adrenocortical theory is correct, high levels of fear should accompany poor

retention which is inconsistent with the results of the present study.

It may be the case, however, that both the parasympathetic nervous

system and the adrenal system (or some other hormonal systems) may interact

to mediate cyclical variations in retention. That is, epinephrine and corticosterone

or some other hormone(s) may fluctuate out of phase with each other such that

neither alone can account for all of the experimental findings of variations in reten-

tion, but can operate together to account for cyclical variations in retention

(Holloway and Wansley, 1973b). Testing this hypothetical neuro-hormonal inter-

action would require the direct manipulation of both epinephrine and corticosterone

levels to determine its effect on cyclical retention. A shock-induced neuro-hormonal

interaction hypothesis would be refuted by the demonstration of cyclical retention

when no shock is employed, e.g., tasks based on reinforcement.

In addition to its theoretical import, cyclical variations in retention are

also relevant to methodological issues in animal learning. It suggests that the time

of training and testing may be critical variables which influence experimental re-

sults and if not controlled for appropriately, may be confounding variables. It is

an interesting possibility that the variability or even nonreplicability of many

experiments may be due to inattention to these simple factors.

The demonstration of cyclical variations in retention has so far only been

demonstrated in rats and mice; however, it would be of interest to determine if

periodicities in retention occur in other organisms including man.

Cyclical variations in retention have been demonstrated only in situations involving shock. Shock may function as a Zeitgeber which entrains some psycho-physiological process causing retention to have a cyclical pattern. That is, it is possible that shock may reset some timing mechanism of the organism. This could be examined by administering a second shock at some variable time after training and/or at a constant time interval prior to the test for retention. If shock functions as a Zeitgeber, it is expected that the second shock would reset the organism's timing mechanism and cyclical retention would be different from the results of the present study. Furthermore, the function of shock as the "primary" Zeitgeber would be questioned if cyclical retention were found in tasks that did not involve shock, whether they be aversive (avoidance of a noxious auditory stimulus), or appetitive (instrumental learning for food reinforcement).

It would also be of interest to determine how the activity cycle of the organism affects cyclical retention. Specifically, this would require taking base-line measures on the activity of each subject and then examining training and testing for retention at various points of the activity cycle.

The existence of biological rhythms in animals is well documented (Holubar, 1969; Marler and Hamilton, 1968; Harker, 1958). Almost all animals including man show periodicity in many of their biological functions, the most prominent rhythms being circadian. Holubar (1969) has suggested that some biological rhythms may reflect the adaptation of a species to external cycles of changing stimulus conditions, i.e., light and temperature, so that their origin is primarily in the rhythmic changes of the environment. These rhythms become so

fixed in the individual organism that they can only be altered with great difficulty by changing the external stimulus rhythm. Not only have rhythms been observed in physiological processes, but in behavioral processes as well. Examples include the sleep-wakefulness cycle, the activity cycle, and feeding and drinking cycles. It is possible that learning and memory may have a biological rhythm. Continued research in this area with more precision in controlling environmental periodicities and using other tasks, organisms, and behavioral variables is necessary.

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