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Editorial

Is is for us an honor and a pleasure to welcome all of you to the XIIIth International Symposium of the Fulton Society on "MEMORY".

A wonderful set of speakers and discussants promise us many hours of enlightment and scientific enjoyement.

The topic of this Symposium, the "Memory" is a fascinating one.

The Greek Mythology so rich in its symbolic meanings gives to our mental faculties the patronage of the gods. Not in vain Greek Mythology declares MNENOSYNE, the goddess of MEMORY, Mother of the nine Muses, divinities of the arts and science.

There is not a single creative act in which the memory does not play a fundamental rol̀e.

More than that, it is basic for man's mental life and progress.

There were artists like Degas who didn't paint directly from life.

His famous pictures of ballet dancers, were not executed with models posing in dancing attitudes, but from the memories gathered during his frequent visits to the essays, from the ballet performed on the stage, or from scenes he had witnessed peering through the dressing room of the ballet girls fixing their attires.

He treasured all these details in his mind, and he confessed that painting from memory, added to his work certain vagueness, mystery and fantasy. And wonderful masterpieces were in this way created until a tragic fate deprived him of his sight.

Unable to master again the brush to express through colours his feelings, he turned to his hands, and aided by memory he produced numerous sculptures recreating with his fingers his beloved ballet dancers which he would never be able to see again.

This in not the only instance in which memory saved a genious from despair.

It is worthwhile to remember Beethoven, that giant of music, whose language too rich to be expressed through words, needed the multifarious throats of the symphony to communicate his feelings, ranging from the grandiosity of the cosmic elements, to the softness of the moonlight shimmering on the waters.

Like Degas he suffered the most crushing of the blows. The sonorous life so dear to him was steadily receding into a fog of silence.

Degas' eyes were closed to colours, Beethoven's ears were blocked to sounds.

In the climax of his desperation Beethoven roared, "I will take Fate by the throat". And MNEMOSINE the titanic godess of MEMORY came to his aid.

Isolated from the world by a wall of silence, his musical inspiration could not be halted by destiny; in his memory; sound was still alive and ready to come to his creative command. From his indomitable courage upsurged the miracle.

His extraordinary memory spared him from the lack of external reference.

A mathematic memory incentivated his inventive powers, and elevated the spiritual content of his music.

The Heroic Symphony is considered an heroic expression of his own life.

No creator has been so alone mastering his art. Deafness has elevated the pureness of his music and profundly aided his spiritual development.

The Heroic Symphony, the Solemn Mass, and the Ninth Symphony, emerged from the very depth of his soul as an expression of faith and courage.

Nowadays we are not devoid of examples of artists who have endured adversity and valiantly surpassed their ill fate.

The case we will refer to, deals with imagery and the visual system.

Studies performed by Ronald Finke have demonstrated the close relationship of mental imagery and the visual system.

Imagery it has been affirmed, is able to produce a much richer variety of visual properties that it has been thought. It involves distance, and THE CAPACITY OF MENTALLY PERCEIVING MORE PRECISE DETAILS IF AN OBJECT IS MENTALLY SEEN INA CLOSE UP examination than those imagined at a larger distance.

Imagination is part of a creative memory fed by past experiences mainly from the visual system. Recent findings states that both of the share the same neural process.

As Beethoven creating from memorized sounds, Carolyn James, a painter condemned to blindness by retinitis pigmentosa, achieved an extraordinary feat that surpassed that of Degas who resigned to translate into sculpture what he would not be able to paint again.

Carolyn James, at present 43 years old, was declared blind at the age of twenty one.

A corageous Scotwoman, she was determined as Beethoven to continue creating in spite of her enormous handicap. And she began to paint inspired by her mental images.

It is surprising what memory can achieve. Seated in front of a canvas, Carolyn worked with watercolours, executing wonderful landscapes.

In front of her were lined up twenty four jars whose colours she knew by heart.

To paint a picture she ought to master a directional memory, and to direct the brush to the exact place of the canvas, to mix colours and to foresee their effects. She goes from one sector of the painting to the other, testing the finish of her painting with her fingertips.

Carolyn as Beethoven have shown that sound and colour are not outside us, but gathered and kept by memory in our nind ready to come to the aid of a creative artist isolated from the world.

Memories of events occured in childhood, are for many artists the source of inspiration in their adult life. And that was the case of the encounter of Dante and Beatriz when he was only nine years old. Its emotional impact was the seed which would flourish in the inmortal work "The Divine Comedy" written by this celebrated poet.

Memory is a fascinating subject from the neurobiological and psychological point of view. Neurobiologists aren trying to stablish by which means an intricated neuronal net is capable of generating a memory trace and also to explain the extraordinary amount of information that is gathered through life and kept in the reservoir which is called memory.

One of the main problems consists in stablishing if the structural organization of the memory is determined by a single entity with a single neural substrate or as an entity integrated by several separated sub-systems that are constituted by different memory stores from the functional and anatomical point of view. Considering the dynamic aspects of memory the question is posed to explain if the process which generates it, is unique with a single neuronal substrate or by fractionation of separated but interdependant processes for instances encoding storage and retrieval, sustained on differential neural contributions.

We will refer now to the structural organization of the memory.

It must be also taken into consideration the point of view of the psychologists who try to demonstrate all the changes that may occur in the memory during the course of time, stablishing different types of memories.

How the storage is produced, how is clasified the colective information and in which degree the emotional state, may reinforce memory, making that some events may be remembered in such a clear and vivid way as they were when captured by the neurons.

In medical literature the case of a very distinguished musician is frequently mentioned, who suffered from encephalitis which greatly damaged his memory. He could not recall what he had just eaten or remember past events of his life, neither the songs which he was fond of singing with his friends. However, he was able to sing and to conduct a chorus, which means that his musical ability was notably preserved. This leads us to think that some sections of the archives of the mind store facts, such as names, images, and events; while other sections store procedures implicate in how to do things.

Other studies on the memory deal with the memory duration; there is a short term memory, which is employed in recalling a fact for a few minutes such as a telephone number which is immediately forgotten or retained for a few hours when a special attention is devoted to it. Its average capacity for retention corresponds to five bits of information.

Concerning the long term memory we could say that this may last for several decades of that which has been experienced for a few seconds.

Once embedded in the mind, the memory is eternally retained.

The long term memory may be arranged by subjects. The recalling may be *frequently brought to the mind when a fact with similar meaning is observed. For example, episodes observed during a sports match may be resurrected during a televised major League game.*

The sense of smell may elicit the remembrance of important and far past events.

The hipothesis has been posed that the same brain structures are involved in the reception of smell, basis emotional responses, and long term memory.

The emotional state is an important factor in sustaining long term memory.

Happy remembrances are more apt to come to the mind than the unhappy ones.

When the mind has processed the same events several times the particular one are replaced by the general ones.

Very frequently we do not remember what we have eaten two days before, because this episode has been repeated several times.

However there are persons who are able to remember the details of the meals of every day. A very surprising fact if the one related with the musicians of an orchestra that frequently play by heart the whole score without any mistake and what is really stonishing is the case of the orchestra conductor Arturo Toscanini, who was able to memorize every single note for each instrument belonging to one hundred operas and two hundred and fifty symphonies.

If some of us look at a scene and close our eyes we are able to retain its image for only two minutes.

The persons who have a photographic memory retains this image for a much longer time. This is specially true in children; afterwards, with the years this capability is diminished. Only one per cent of the adults has a protographic memory.

Many persons are plagued with forgetfulness. In some cases the failure is amazingly specific, for example, there exists the possibility that one is unable to recognize faces, which is prosopagnosia, that is to say difficulty in recognizing familiar faces.

It is admiteed that the memory of a person includes a template of his face, according to the changes observed in the EEG when the person is confronted with the figure of a relative or recognized celebrities.

Seeing the face, activites the template that elicits the array of interneuronal impulses that aim to memorization. In some instances these electrophysiological rhytms are immediately arrested after the necessary information to recognize the face, to find the word, or to remember the happening annexed to the image.

Regarding the studies of memory concerning the sectors in which the brain forms an stores it, notable progresses has been attained.

The structures that are implicated are the hypocampus, the amygdala, the diencephalum, the cerebral cortex and the cerebellum. The dentated and interpositus nucleis of the cerebellum are involved in the storage and organization or stimulus response associations (Mc Cormick and Thompson).

As Mishkin affirms, integrated perceptions are anchored in two structures on the inner surface of the temporal lobe in both hemispheres of the brain: the hipocampus and the amygdala. By damaging both structures at once, in both brain hemispheres of test monkeys on observed in the animal a gloval anterograde amnesia.

On recognize that the amygdala it is the structure largely responsible for adding a positive association to a stimulus processed by the visual system.

The diencephalic nuclei are implicated in memory. The diencephalum receives fibers running from the hippocampus and the amygdala.

The likeliest repositories of memory are the same areas of the cortex where sensory expressions take shape. The subcortical memory circuits must therefore engage ina kind of feedback with the cortex.

Suffering amnesia for events it is believed to be caused by damage to the hippocampus. It is supposed that in the hyppocampus are processed new facts and sent for storage elsewhere.

Researchers led by Stuart, Zola-Morgan of U.C. San Diego have informed that they have defined a layer of 4.6 millions cells in the hyppocampus where new facts memories are processed.

In rats with hippocampal lesions there are profund deficits in tasks that require memory for spatial location (Kesner and DiMattia-Olton).

In Olton studies damage to input and output pathways from the hippocampus including medial septum, fimbria fornix and entorhinal cortex produced the same kind of impairment.

On the other hand, the connections that are in the brain by which means memory is processed is being investigated.

The afflux of memories sometimes is elicited by smelling a perfum. The olfatory nerve conveys scens information to the hippocampus.

It is accepted that the hippocampus and the neighboring structures of the brain are responsible for eliciting emotions.

This puts in evidence the fact that feelings are important in creating and maintaining memories.

These structures are important as much for memory as for emotion, said Mortimer Mishkin.

From the hyppocampus, fact memories probably travel to the cerebral cortex

In this region simple memories are located to persist in special areas that correspond to the senses. However, up to now it has been impossible to determine where the memory is kept concerning a complex happening as a novel scene.

It is accepted that which is going to be memorized arrives at the cortex through one of the senses and it is analysed. This information is oriented to the hippocampus, finally into the depth of the brain. Chemical changes begin to occur and in some mysterious way they preserve the trail of memory.

The neurons all along the pathways of information on the brain may be forever modified and in this way to preserve the details of a paintings. Seeing again the same painting activates the trace and the memory.

We will mention now studies performed on the synapsis, connections are vital to trace formation. Comparing a group of rats that were kept in a complex environment, with another group that were located in empty cages, the rats that were in the complex environment after examining their brain showed a notably increased number of synapsis in the cortical sectors compared with those which were kept in isolated cages.

This leads to the conclusion that the synapsis which is formed as in the case of the rats process information are seats of memory.

The neurons form the synapsis that might constitute memory. When an electrical impulses throbs through the neurons, which happens when the information arrives, calpain enzime located in the dendrites facilitates the chemical changes so that its membrane is elongated and reaches neighboring neurons and form synapsis with them, which might form the first link of the memory trace.

If the neuronal changes give place to memory, the question is posed which is what determines that something that is memorized is kept for decades while other remembrances disappear. Eric Kandel and collaborators from Columbia University think that the explanation can be found in the genes, the molecules of heredity. It has been suggested that if incoming memory turns certain genes on and others off, these changes in activity may in some way elicit permanent changes in the neurons and this may be the basical fact of permanent memory.

Genes which contain the blueprint for our bodies. if they are well provided of specifications for our mind, may be thus the carriers of memory.

Ojemann, Creutzfeldt and Lehich, have profited the cases of medically intractable epilepsy in which are performed resective surgery under local anesthesia to investigate basic cortical mechanisms of human higher functions. Are observing changes in neuronal activity related to language and memory as recorded from temporal cortex of the dominant hemisphere while resective surgery is performed under local anesthesia.

Neuronal recording has been done utilizing special microelectrodes and its output was amplified and recorded through an appropriate instrument.

Recordings generally contained the activity of single neurons or activities from two or more cells easily distinguished by action potential amplitude.

By these studies about what happens on neuronal activity in lateral temporal cortex of the dominant hemisphere during naming or memory, input has put in evidence different relationships. Neuronal populations that are entailed to both naming and memory output, has been observed.

The frequent identification of lateral cortical neuronal populations related to memory input is an evidence of the rol the lateral temporal cortex in short term verbal memory mechanisms. These neuronal populations provides the link between language and recent memory.

The narrow proximity of the basal temporal language area, with the hyppocampal gyrus lead us to think that its function may be closely related to memory of verbal material.

Rausch and Babb also profiting the possibilities of research that the surgery of epilepsy offers, have studied memory specialization within the mesial temporal lobe in man.

It is well known that the anterior temporal lobe in man is implicated in memory processing. The bilateral removal of the temporal lobes generates a severe amnesia syndrome, but if the resection is done only on one of the temporal lobes, this originates an impairment in learning of either verbal or non verbal material, which depends on the implicated hemisphere. These findings were acquired largely from epileptic patients that have suffered resection of the temporal lobe with the purpose of eliminating an epileptic focus. The indication of one potentional functional diferentiation into one temporal lobe has been deduced from correlations of the degree of memory deficit with the extension of the ablation of primarily mesial versus lateral neocortical regions. The loss of the mesial temporal lobe in the non dominant hemisphere in which amigdala, pes hipocampi, gyrus hipocampi and uncus are involved, it is considered to be related with impairment in learning of simple spatial material.

Instead the damage to the lateral neocortical areas produces deficits in memory for abstract visual patterns. The resection performed of the language dominant temporal lobe has not had the same difference. On most tasks the resultant verbal memory defficit is related to the extension of removal of mesial and lateral temporal lobe areas.

These surgical resections have not demonstrated differentiation of subregions within the temporal lobe. This study gives one significative analysis of anatomical regions of the left temporal lobe that relates to simple versus complex verbal learning.

In the mentiones study the patients suffered from a left temporal lobe epilepsy associated with different degrees of verbal memory deficits.

It has been observed that the left temporal lobe is specific to the verbal memory tests and the right temporal lobe is specific to the visual non verbal memory tests.

As a conclusion of the important research of Drs. Rausch and Babb the suggestion can be accepted that the mesial temporal lobe regions are specialized in different types of memory processing. The sectors anatomically more simple, prosubiculum, smaller, thin layered pyramidal cells seem to be more implicated in learning of simple rote verbal associations, while the complex, multi-layered gyrus hippocampi is more involved in complex, semantic verbal learning.

If we consider that the widespread projections of the gyrus hippocampi to almost all association cortices, it is not surprising that this area is very much involved in the processing of linguistically complex material.

It is amazing the negative correlation of subicular cell density with complex *verbal learning, which is different in the gyrus hippocampi the neuronal density of the subiculum has not a significative correlation with that of the pes hipocampi, the region more prone to lose cells in this patient population. The general integrity or the pathology of the mesial temporal lobe is not related to the number of neurons in the subiculum. The fact that the loss of subicular cells is not related to impaired complex verbal learning, suggests that this transitional cortex is not essential for semantic memory.*

We will refer now to studies performed by J. A. Walker, R. G. Stanoulis and KD. Laxer about what stage of memory do the temporal lobes serve A sodium amytal perspective.

The intracarotid sodium amytal test offers an excellent opportunity to establish and evaluate hypothesis about the nature of memory deficits in patients with temporal lobe epilepsy.

The intracarothid sodium amytal test permits us to evaluate the capacity of each hemisphere in momentary isolation from the other hemisphere and in these conditions the exploration about the nature of the underlying memory deficit may be employing establishing a direct comparison between the two hemispheres, the one which is in deficit and the one which remains intact.

With these studies it is suggested that two different forms of amnesia exist one associated with diencephalic damage and difficulty in encoding information and one related with limbic system damage and difficulty in post-encoding processes.

Korsakoff amnesics are poor in encoding input. Amnesics with limbic systems lesions are impaired in storage. In cases where an extensive cortical lesion exists, such is in encephalitis cases or perhaps Alzeimer Diseases, a marked impairment in retrieval of very old information as well as in acquiring new information, is observed which may constitute another type of amnesia.

The intracarotidial sodium amytal test is a technique that allows to evaluate the competence of each hemisphere in relative isolation from the other.

We will refere now to studies performed by Gregor W. Jason about manual learning and performance after surgical excisions for the control of epilepsy.

Studying the results obtained from the Copy Single Position and the Metronome Paced Performance task the authors found a difference between remembering and copying manual tasks.

Left and right hemisphere lesions produced similar effects. On the contrary in the manual sequence Learning task in which remembering was emphasized. left side lesions (performed on the frontal or temporal lobe) determined a deficit while the same lesions (done in right sided of frontal or temporal lobe) did not show the same result causing no deficits.

The site of the lesions within the hemisphere have significative results, the left frontal and the left temporal lobe have demonstrated that they are implicated in learning series of discrete hand positions.

There was a general effect of some of the lesions on the copying tasks this *was probably originated by the ample variety of demands in the tasks related to quickly analyzing the demonstrate positions, determining the necessary movements to imitate the position, performing the movements and giving a close attention to all the details involved.*

There was also a notable deficit of both frontal lobe groups on the Metronome Paced Performance task, suggesting a role of frontal cortex of either side in performing multiple positions quickly. This happens in both contralateral and ipsilateral hands and shows an interesting parallel concerning studies, exhibiting an increase of blood flow in the suplementary motor areas of the frontal lobes when either hand is performing a sequential finger-tapping task. However there was not a clear evidence for the specificity of suplementary motor areas lesion in this group due to the small number of cases involved in these studies, it is not possible to obtain a clear answer to the question.

The precess of memory has been studied in the different stages of life. We will refer to "Infantile Amnesia" it concerns those cases in which the person can not remember facts occurred when they were three years old. This happens because the brain structures involved in declarative memory does not reach its complete development until the child reaches one and a half or two years of age. It has been thought that the hippocampus is one of the brain structures that demands more time to become fully functional, and until this is not attained, the child can not permanently record fact memories.

Besides, in this there is also implicated a fact of psychological meaning, the child has a perception of the world different from that of the adults. The Child is devoid of the sense for detecting what is usual and what is unusual.

Generally he can't remember important events in the family life, such as the birth of a brother. And also when his mother left home to go to a sanatory to give birth to a child or her return with the baby. because he can not differentiate what is trascendental or not in daily life.

It is more common that the child remember facts derived from the rutinary and habitual and gives them a distinctive meaning. That is to say, that the importance of the event to be remembered is not significative.

The adult mind which has reached maturity is based upon words, which the child lacks. Events lived in infancy not apprehended by the corresponding word, were not remembered.

Memory begins to deteriorate in the third age. It is believed that the 85 % of the adults in perfect health, those who are more than 65 years old, show some symptom of memory deterioration. When a person grows in age an increase of information has been acquired and retained; but the percentage that can be remembered without difficulty, remains the same, in a certain degree he is forgetting more.

It is very frequent the fact that there is a diminution of the ability to remember the name of a person recently introduced. Another important fact is that the person who in his youth had difficulty to remember a number or a list of things has this deffect increased in old age.

Short-term memory doesn't become less complete or accurate, though it may get slower.

Long term memory is more persistent because it has had more time to consolidate and to correlate with many locations in the cortex. Besides, old memories are also retained because they are often remembered. A memory frequently revived lasts longer. The old person tries to maintain his memory to keep and retain access to important things.

These exists the possibility that memory deficits are due to the fact that old persons are smarter in forgetting what is unimportant to them. It is believed that the ability to forget what has been previously memorized allows old adults to focus on underlying principles.

We will refer now about the effects of alcohol on memory. Even by drinking moderately memory is affected. Drinks act as depressors which make the person to be less alert and thus he is unable to remember what happens under its influence. Besides alcohol acts on the brain areas where memory is elaborated. It has been demonstrated that interferes more in the stage of consolidation of information into long term memory than with the retrieval of it.

Alcohol acts on the hippocampus and in this way interferes with the organization of new memories.

Excessive drinking may damage the memory permanently.

Regarding marihuana it affects memory. It inhibits the transfer from short to long term memory, but leaves existing long term memory intact. It is affirmed that once the information gest into our memory, the retrieval of it is less disturbed by the marihuana.

Marihuana affects the enconding of new information which constitute a vital step to filing it in long term storage.

On the other hand, stimulants may produce a sharp memory because it isustains the alertness. Experiments performed in rats shows that the memory is increased with the use of drugs that allows more exygen reach the brain and this is probably the consequence that the animals are more alert, without being mediated in a direct way an improvement of the memory.

Concerning investigations of products that increase memory, it has been observed that rats that receive soybean oil are more productive in its memory. The fatty membranes of neurons implicated in memory harden with age but polyunsaturated fatty acid keeps neurons pliable and are more efficient to carry our nerve signals and thus to preserve memory.

Considerable progress has been attained in recent years to the comprehension of such an important subject as memory.

New vistas are opened that promise us a better future for humanity because the knowledge adquired about memory will enable men of science to enhance it, to prevent it loss and to repair its damage.

Prof. Dr. VICTOR SORIANO

Anatomical Substrates of Interictal Memory Deficits in Temporal Lobe Epileptics

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INTRODUCTION

It is well-known that patients with temporal lobe epilepsy demonstrate varying degrees and types of memory disturbances. There is a selectivity to their memory problems that correlates with the anatomical locus or laterality of the seizure. Patients with left temporal lobe epilepsy are more likely to have problems with verbal learning, while patients with right temporal lobe epilepsy have more difficulties with learning nonverbal material (Delaney et al., 1980; Milner, 1972; 1974; Rausch, 1985). These selective deficits are usually enhanced by unilateral resection of the involved temporal lobe (Milner, 1974; Rausch and Crandall, 1982).

Our research has concentrated upon the contribution of an underlying pathological substrate of the epilepsy to these expressed memory deficits. A common pathological finding in temporal lobe epilepsy, and in particular temporal lobe epilepsy with unknown etiology, is hippocampal sclerosis or loss of neurons in the affected hippocampus (Babb and Brown, 1987). The cell loss tends to be restrictive and does not involve the overlying neocortex. These selective natural-occuring lesions provide un unique opportunity to study brain-behavior correlates in humans without complications due to other pathologies or larger excisions of the temporal lobe. We have been able to relate parameters, such as extent and pattern, of these focal hippocampal lesions to normal and abnormal memory functioning.

METHODOLOGY

PATIENTS:

Those patients which have made these research endeavors possible are intractable epileptics whose focal neurological condition results in highly selective lesions within the temporal lobe. These patients typically have no known etiology for their epilepsy and have no radiologically-diagnosed structural lesions. These patients have complex partial seizures which have been documented, primarily by depth electrodes (Crandall et al. 1983; Engel et al., 1983) to originate from the mesial temporal lobe. Laterality and focality of the neurological involvement are made by extensive medical work-up which includes simultaneously-obtained video and EEG recordings of spontaneous seizures as

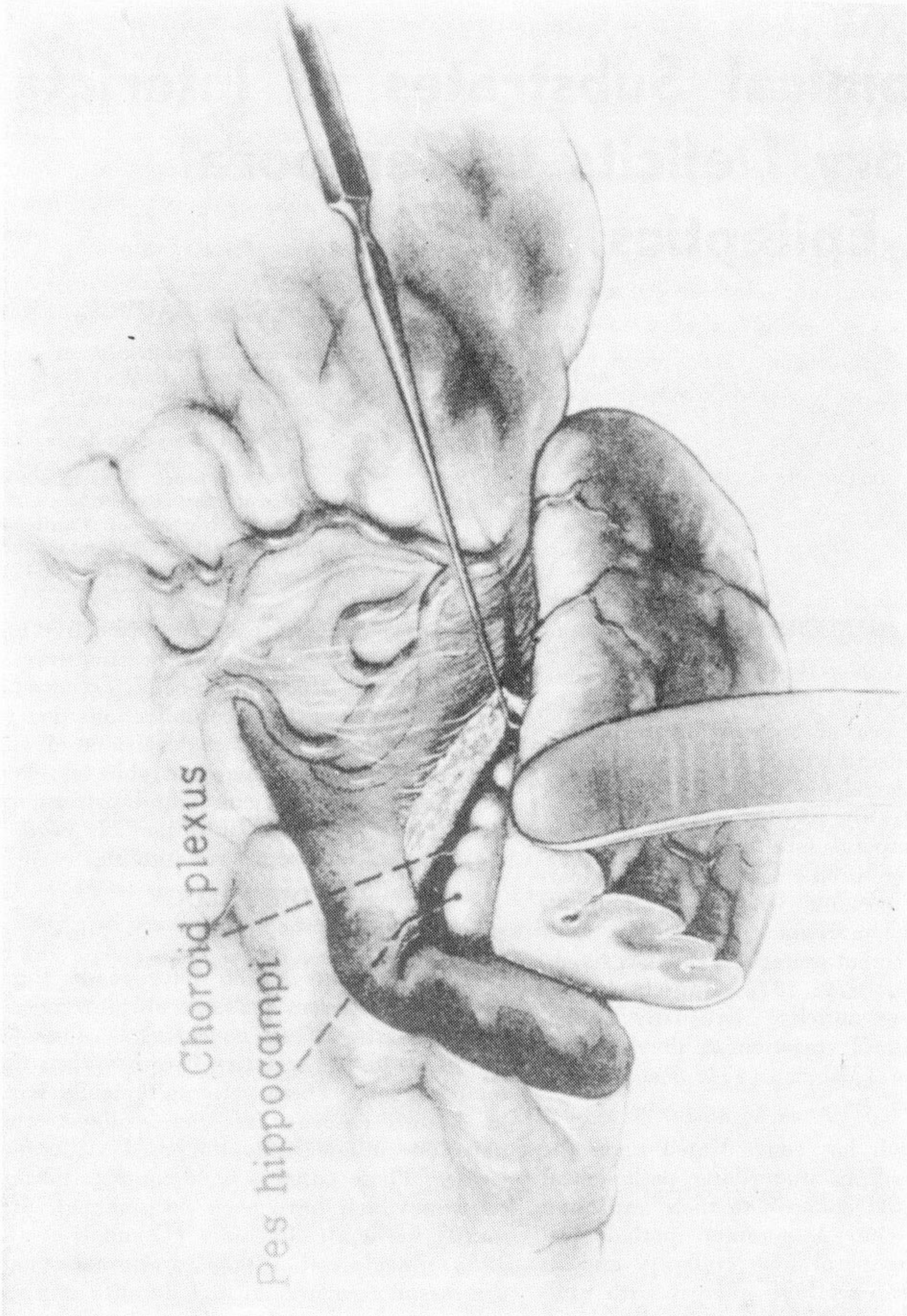


Fig. 1. — *Schematic of the en bloc resection of the anterior temporal lobe performed by Paul H. Crandall (From Brown, 1973; reprinted with permission).*

well as cerebral functional tests, such as positron emission computed tomography (cf., Engel et al., 1982a; 1982b) and neuropsychological examination and intracarotid Sodium Amytal procedure (cf., Rausch, 1987; Rausch and Risinger, 1988). Because of the cerebral focality of their neurological disease, the patients typically have average or above average intelligence. Except for memory functions, their cognitive skills are not significantly affected. For example, cognitive domains, such as attention, concentration, language, visual-perceptual and motor skills, fall within normal limits. It is because of the intactness of most cognitive skills that we are able to clearly delineate the specific memory deficits in these patients.

SURGERY:

We have been able to study the hippocampal damage in these patients because of the tissue available from the surgery performed to treat their intractable epilepsy. The surgical technique used by Dr. Paul Crandall is a stardard *en bloc* resection of the affected temporal lobe (Crandall, 1987). The surgery (shown in Figure 1) removes 3 cm of the hippocampus as well as the hippocampal gyrus, the lateral 2/3's of the amygdala, the uncus, and the lateral neocortex. The tissue is removed *en bloc*, fixed, and processed for light microscopy. The lateral neocortical resection is slightly smaller on the left temporal lobe (5.5 cm) than on the right temporal lobe (6.0 cm). The standardization of the surgery assures the anatomical representation of the tissue available for analyses. It also allows for controlled conditions in which to compare surgical effects of various patient groups.

PATHOLOGY:

The degree and pattern of hippocampal sclerosis found in the resected tissue may vary markedly across patients, with hippocampal cell loss ranging from minimal to severe. Also, while certain subregions of the hippocampus tend to be involved, there exist varying patterns of damage among patients (Babb et al 1984; Babb and Brown, 1987). Some patients show cell loss of selective regions, while others show involvement of multiple subregions. Damage along the anterior-posterior axis may also vary. For example, some patients exhibit marked hippocampal damage throughout the hippocampus, while others may show marked damage limited to either the anterior or the position section. These variations in hippocampal damage allow for correlative studies with the differences observed in memory functions.

The extent and pattern of the hippocampal damage are quantified on the resected tissue by calculating the neuronal cell density of the regions of the temporal lobe. Volumetric cell densities are computed by counting the number of somata (pyramidal cells) or nuclei (granule cells) using a grid-sampling procedure (Babb et al., 1984). The neuronal counts are corrected for areal density and then corrected for volumetric denisity using the Abercrombie (1946) corrections for section thickness. The following anatomical regions, based on the nomenclature of Crosby et al. (1962), are routinely analyzed (see Figure 2): fascia dentata, upper and lower, granule cells; hippocampus (CA4, CA3, CA2, CA1 and prosubiculum); subicular complex (subiculum and presubiculum); hippocampal gyrus; fusiform gyrus; inferior and middle temporal lobe gyri. (Refer to Figure 2). Cytologic analyses of the amygdala are not routinely performed, although observations of the amygdala's integrity are made at surgery.

NEUROPSYCHOLOGICAL MEASURES:

Psychological data have been obtained from standardized neuropsychological evaluation or specialized research batteries administered to the patients prior to and one year following surgical resection of the temporal lobe. The standard evaluation measures a broad range of cognitive domains including, but not limited to, intelligence, language, visual-perceptual and visual-constructional skills, attention and concentration, and verbal and nonverbal memory functioning (cf., Rausch, 1987). Administrations of the various tasks are standardized and the results can be quantified. Normative data are available from a number of

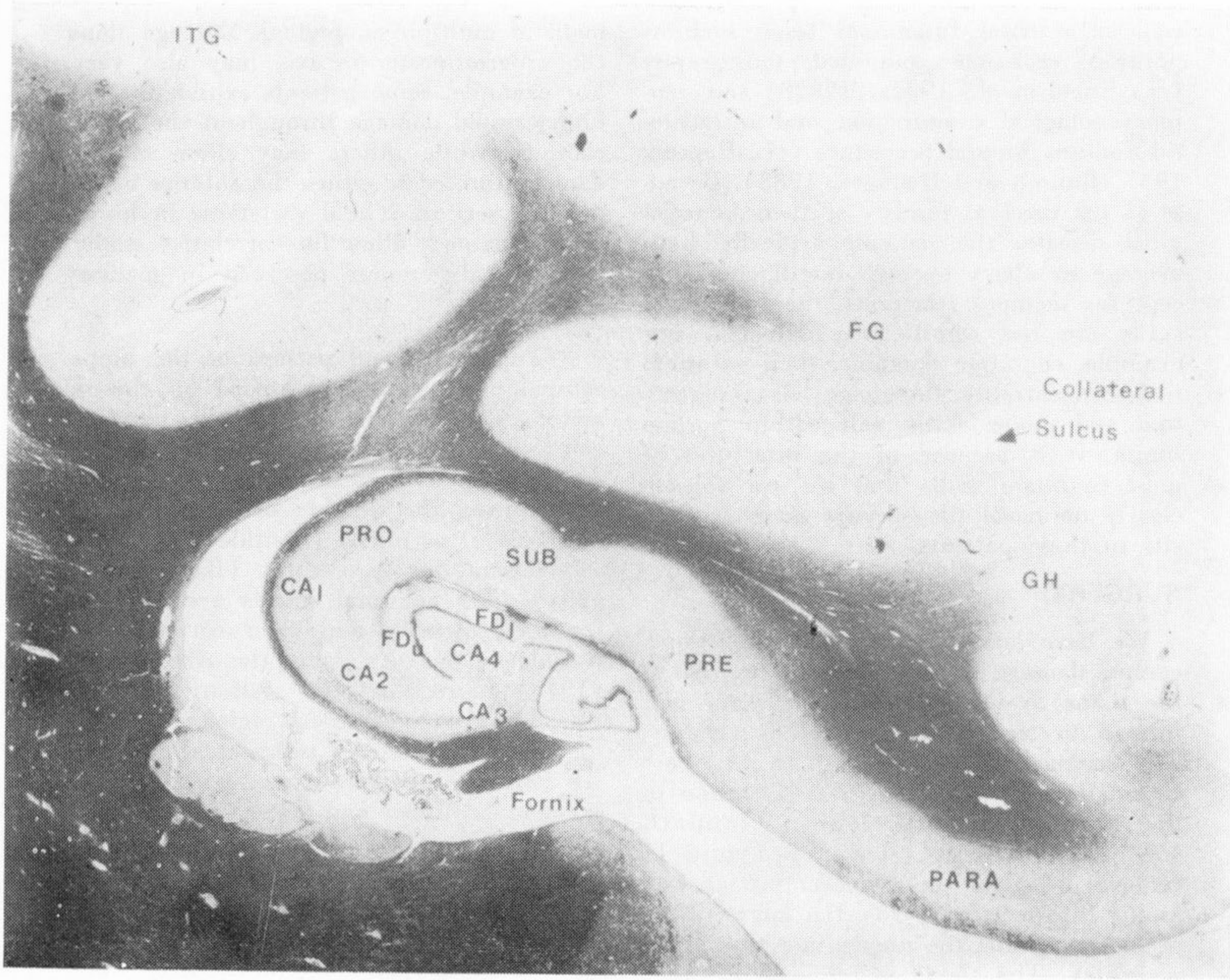


Fig. 2. — *Photomicrograph of a section from a normal temporal lobe. Regions routinely analyzed for volumetric cell desities are: fascia dentata, upper and lower (FD_1 and FDu); hippocampus (CA4, CA3, CA2, CA1, and prosubiculum [PRO]); subiculum (SUB); presubiculum (PRE); hippocampal gyrus (GH); fusiform gyrus (FG); inferior temporal gyrus (ITG); and (not shown) middle temporal gyrus.*

published sources, including our own normative data base (Rausch, 1986).

The psychological scores obtained preoperatively allow for correlation with the underlying pathological changes as well as provide a baseline from which to evaluate postoperative cognitive changes. The postoperative psychological scores are used to evaluate change in the individual patient as well as to confirm the role of temporal lobe structures in selective cognitive processes. The postoperative test scores are also useful in determining the differential effects of left and right temporal lobe lesions.

In addition to the neuropsychological tests administered, patients undergo the intracarotid Sodium Amytal procedure (Wada and Rasmussen, 1960) for prognostic and diagnostic reasons (Rausch and Risinger, 1988). Information is obtained as to the hemisphere dominant for language as well as hemispheric memory competence. Knowledge as to the hemisphere dominant for language is important for identifying homogeneous patient groups.

FINDINGS

CORRELATION OF SELECTIVE MEMORY LOSS WITH LATERALITY OF TEMPORAL LOBE LESIONS:

Numerous reports have indicated that resection of the left temporal lobe results in significant verbal memory deficits, while resection of the right temporal lobe results in difficulty learning material not easily verbalized (Milner, 1972: Milner, 1975; Kimura, 1963; Taylor, 1969). In our patient population, we have found a similar differential effect of laterality of the lesion (cf., Rausch, 1985). However, the correlation of verbal memory deficits with left (language-dominant) temporal lobe resections is stronger than the correlation of nonverbal memory deficits with right temporal lobe resections, despite the larger neocortical resection of the latter patient group (cf., Rausch and Babb, 1987). The source of the relative weakness of correlative behavioral changes with right temporal lobe resections is unknown. It may be that we have not identified a behavioral measure which most clearly taps right hemisphere functions. Or it may be that functional dependency upon neural structures in the right temporal lobe is not as closely associated as those in the left temporal lobe. This latter interpretation is consistent with the conclusions of others that there is a more diffuse representation of functions in the right hemisphere than in the left hemisphere (De Renzi and Faglione, 1967; Kertesz and Dobrowolski, 1981; Semmes, 1968). Also, it may be that the lack of strong correlations in our right-sided patient group reflects the fact that the right temporal lobe group is a more heterogeneous group of patients than our studied left temporal lobe patients. Research designs are usually restricted to those patients with left hemisphere language dominance. Left temporal lobe patients whose language has pathologically shifted to the right hemisphere, as determined by the ISA procedure, are excluded from typical analyses (Rausch and Walsh, 1984). As yet, we are unable with our current ISA procedure to identify those right temporal lobe patients whose nondominant functions may have shifted and consequently co-exist with verbal functions in the left hemisphere. Therefore, because of the strong and consistent correlation of behavior changes with left (language-dominant) temporal lobe dysfunction, we have concentrated our initial investigations on patients left (language-dominant) temporal lobe epilepsy.

CORRELATION OF HIPPOCAMPAL (Ammon's Horn) CELL LOSS WITH POSTOPERATIVE MEMORY LOSS:

The region of CA4-CA1 or Ammon's horn of the temporal lobe is highly sensitive to cell loss in this temporal lobe epileptic population (Babb and Brown, 1987). The area also plays a significant role in verbal memory functioning. In a study of the change in overall verbal memory functioning following left temporal lobe surgery, the degree of memory loss was found to correlate with the integrity of Ammon's Horn (Rausch et al., 1984). (Refer to Figure 3).

Those patients with high volumetric cell densities of the left hippocampal subregions showed the greatest decrease in a composite verbal memory factor score following resection of the left temporal lobe, indicating that these neurons had been maintaining this memory function. Left temporal lobe epileptics with low cell counts of the hippocampus did not show the same degree of memory loss, suggesting that the neuronallydeficient hippocampus preoperatively was not involved in the behavior. Neuronal density of the right hippocampus in right temporal lobe epileptics was not related to a postoperative change in verbal memory. This latter finding reflects the specificity of the left hippocampus in verbal memory.

CORRELATION OF SELECTIVE HIPPOCAMPAL DAMAGE AND DIFFERENTIAL MEMORY CHANGES:

Not only change in memory functioning after surgery, but also preoperative memory functioning has been related to the integrity of the hippocampus in these patients. The volumetric cells densities of subregions of the left temporal lobe were correlated with different verbal memory measures (Rausch and Babb, 1987). The subregions analyzed included: fascia dentata lower and upper, CA4, CA3, CA2, CA1, prosubiculum, subiculum, presubiculum, hippocampal gyrus, and the lateral temporal gyri. The two verbal memory tests were 1) a simple, rote association taskthe unrelated word-pairs from the Wechsler Memory Sca-

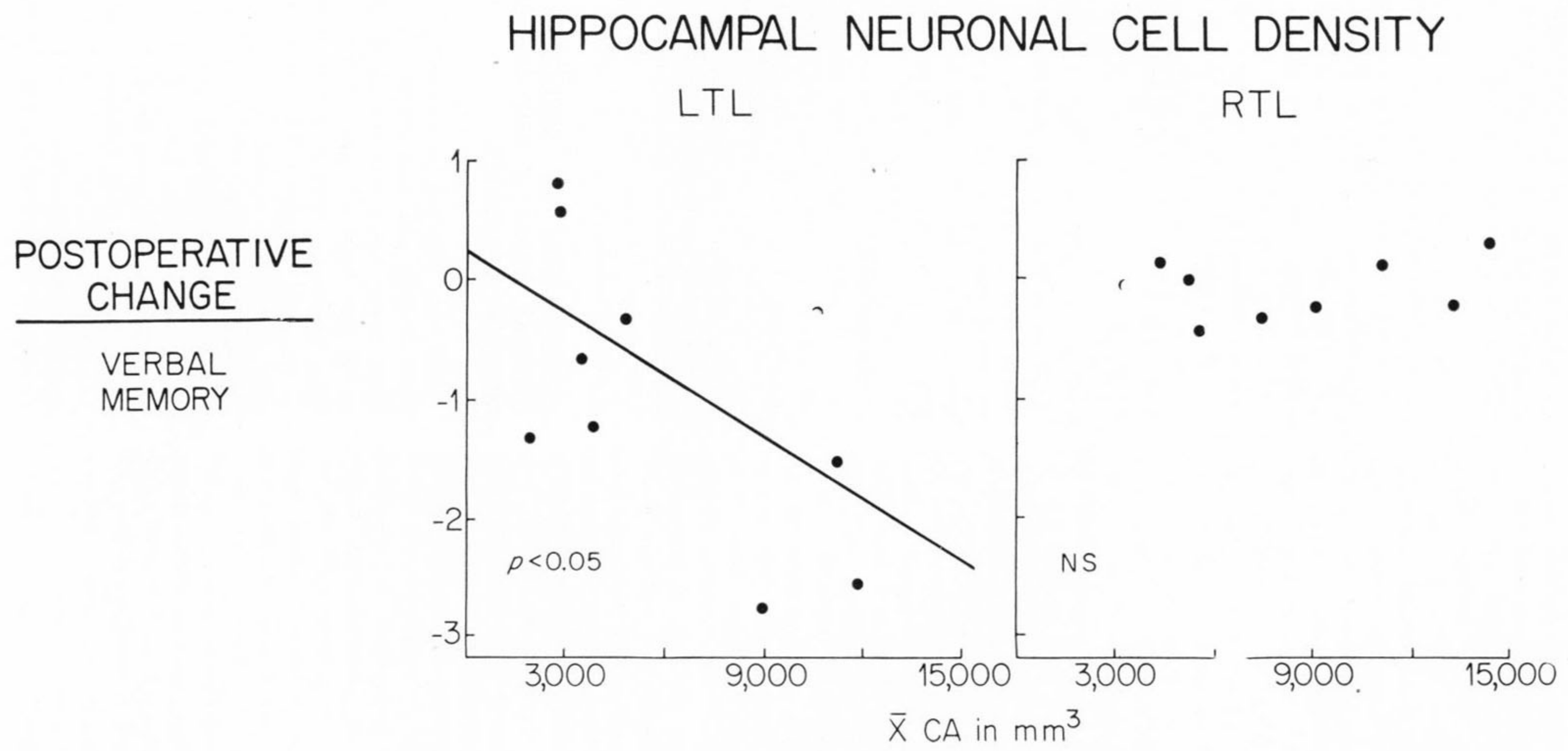


Fig. 3. — *Change in verbal memory factor score following temporal lobe resection is significantly related to the hippocampal cell density in left temporal lobe patients but not in right temporal lobe patinents.*
(Verbal memory factor score was derived by maximum likelihood factor analysis with direct oblimin rotation of scores of 87 epileptic patients).

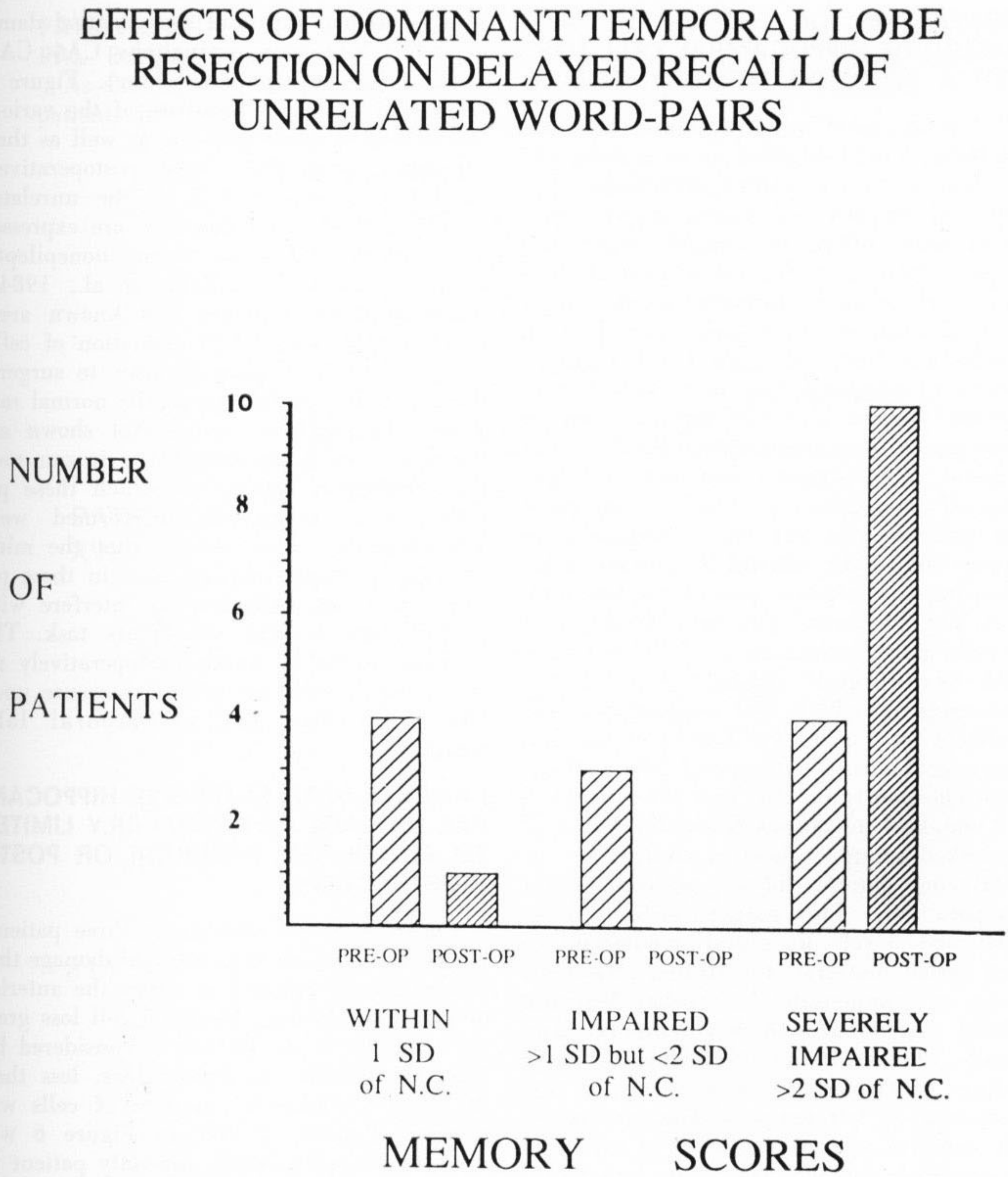


Fig. 4. — *Performance of eleven patients before and after standard left temporal lobe resections. Scores are derived from the 45 minute-delayed recall of unrelated word-pairs, adapted from the WMS. Scores are shown relative to normal controls (N.C.).*

le (WMS; Wechsler and Stone, 1945) and 2) a semantically more complex task, the Logical Prose Subtest of the WMS. Loss of neurons in Ammon's Horn, in particular CA1, significantly correlated with poor performance on the simple, rote association task, including both the immediate and 45 minutedelayed recall measures. Logical prose did not significantly correlate with the integrity of CA1, or any of the other Ammon's Horn subfields. These findings not only confirmed our previous finding of the importance of Ammon's Horn in verbal memory, but also suggested that the integrity of this anatomcial area was directly related to a specific verbal memory task, unrelated word-pairs.

CORRELATION OF THE EXTENT AND PATTERN OF HIPPOCAMPAL CELL LOSS WITH A SELECTIVE MEMORY DEFICIT:

Our most recent series of studies (Rausch and Babb, 1988) enabled us to address the question as to the extent and pattern of structural hippocampal damage necessary to impair performance on a highly dependent temporal lobe task. We found that the delayed recall score of unrelated word-pairs to be consistently and markedly impaired in patients who had undergone left (language-dominant) temporal lobe resections. Group analyses showed a significant decrease in performance from preoperative levels in left temporal lobe patients, but not in right temporal lobe patients. The left temporal lobe patients did not show postoperative impairment on the nonverbal memory tasks. indicating the selective role of the left temporal lobe in verbal memory. Analyses of the individual performances of the left temporal lobe surgical patients revealed the consistency in which this measure was depressed. (See Figure 4). Ten of eleven left (language-dominant) temporal lobe patients performed poorly on this task after surgery, > 2 standard deviations below the mean of a matched group of normal controls.

Only one patient did not perform poorly postoperatively. This patient utilized a sophisticated level of visual mediation to learn verbal material; the strategy has been shown to compensate for verbal learning deficits associated with left temporal lobe damage (Jones, 1974). Nevertheless, these findings confirmed the dependency of this measure upon left temporal lobe structures.

In order to address the effect of extent of hippocampal damage upon this behavioral measure, patients had to have available for microscopic studies resected tissue from the most anterior and posterior sections of the left (language-dominant) hippocampus. Eleven patients, with pre —and one— year postoperative neuropsychological data, constituted the subject group.

PATIENTS WITH MINIMAL HIPPOCAMPAL DAMAGE:

The first subgroup of patients consisted of two patients with left temporal lobe epilepsy who had minimal hippocampal damage. (The hippocampus includes CA4, CA3, CA2, CA1 and prosubiculum). Figure 5 shows the neuronal densities of the various subregions of these patients as well as their memory scores pre —and postoperatively on the delayed— recall of the unrelated word-pairs. The cell densities are expressed as percent reduction from nonepileptic temporal lobes (cf., Babb et al., 1984). None of these patients had known areas with greater than 50 % reduction of cells.

As shown in Figure 5, prior to surgery, these patients performed in the normal range on this memory score. Not shown are the other verbal memory tasks in our neuropsychological battery on which these patients also consistently performed well. Therefore, we must assume that the minimal hippocampal damage seen in these patients was not sufficient to interfere with performance on the word-pairs task. The decrease in performance postoperatively reflects the dependency of this measure upon the left (dominant) temporal lobe structures.

PATIENTS WITH EXTENSIVE HIPPOCAMPAL DAMAGE BUT PRIMARILY LIMITED TO EITHER THE ANTERIOR OR POSTERIOR SECTION:

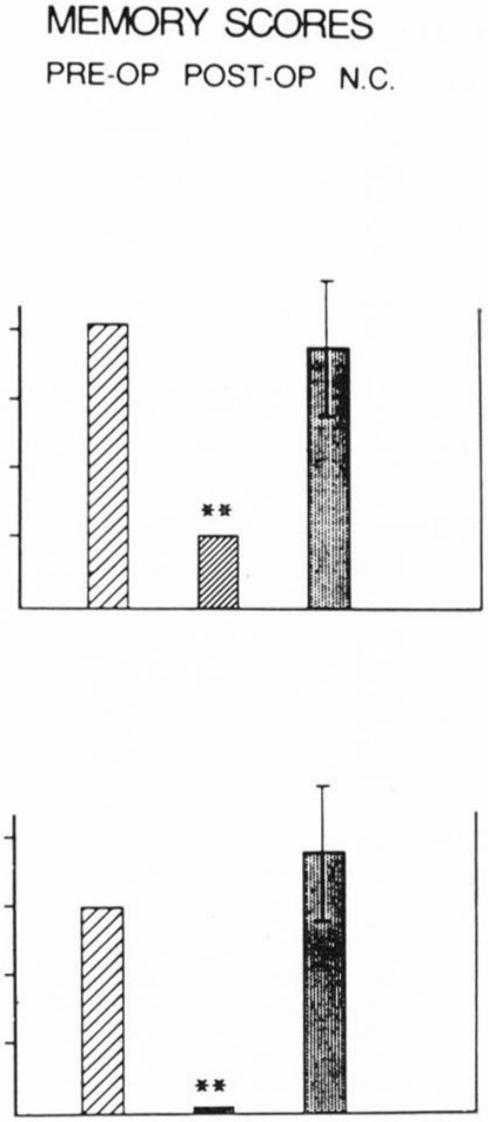
Figure 6 shows data from three patients who had extensive hippocampal damage that was primarily limited to either the anterior or posterior section. Regional cell loss greater than 80 % of controls is considered highly significant. In these areas, less than 20 % of the normal number of cells was present. Patient #XIII in Figure 6 was our exceptional patient; the only patient in these analyses that performed well on this task after surgery. Patient #209 and Patient #181, with extensive posterior hippocampal damage, performed reasonably well before surgery. Thus, these findings would suggest that extensive damage more limited to the anterior or posterior hippocampus is not sufficient to severely impair performance on the word-pairs task. The drop in performance postoperatively in the latter two patients again confirms that the anterior left temporal lobe played an important role in this task.

PATIENTS WITH EXTENSIVE HIPPOCAMPAL DAMAGE:

As shown in Figure 7, the next group of patients had extensive damage of the hippocampal regions in both the anterior and posterior sections. At least three hippocampal regions in each section had 80 % or greater cell loss. Several regions from Pa-

PATIENTS WITH MINIMAL HIPPOCAMPAL DAMAGE

PATIENT #		ANT	POST
XVI			
	FDU	23	46
	FDL	15	22
	CA4	30	45
	CA3	30	41
	CA2	13	40
	CA1	18	41
	PRO	3	33
	SUB	15	25
	PRE	0	0
211			
	FDU	14	29
	FDL	32	15
	CA4	39	10
	CA3	49	-
	CA2	-	-
	CA1	-	-
	PRO	-	33
	SUB	0	18
	PRE	-	5



** > 2 SD below normal controls

Fig. 5. — *Neuron loss (expressed as percent reduction from normal) of temporal lobe regions in two patients with minimal hippocampal damage. Patients' scores in the delayed-recall of unrelated word-pairs obtained before (pre-op) and after (post-op) standard left temporal lobe resections. Normal control (N.C.) scores (with S.D.) are also shown. Data shown in Figures 5, 6, 7, and 8 are adapted from Rausch and Babb (1988).*

PATIENTS WITH EXTENSIVE DAMAGE PRIMARILY LIMITED TO EITHER ANTERIOR OR POSTERIOR SECTIONS

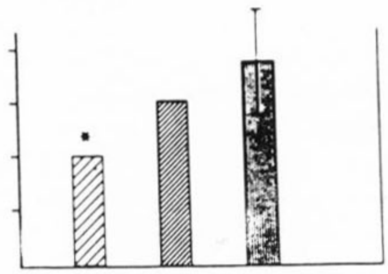
MEMORY SCORES
PRE-OP POST-OP N.C.

ANT POST

PRIMARILY ANT DAMAGE

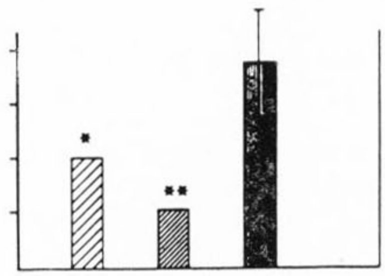
PATIENT #

XIII	ANT	POST
FDU	58	63
FDL	76	58
CA4	94	78
CA3	97	61
CA2	71	54
CA1	87	53
PRO	63	86
SUB	18	45
PRE	0	0

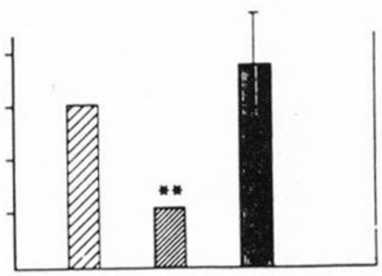


PRIMARILY POST DAMAGE

209	ANT	POST
FDU	64	74
FDL	70	75
CA4	75	90
CA3	44	55
CA2	71	100
CA1	70	83
PRO	66	60
SUB	12	-
PRE	-	-



181	ANT	POST
FDU	62	67
FDL	73	77
CA4	0	80
CA3	97	83
CA2	89	56
CA1	77	100
PRO	48	100
SUB	34	-
PRE	-	-



* > 1 SD below normal controls
** > 2 SD below normal controls

Fig. 6. — *Neuron loss (expressed as percent reduction from normal) of three left temporal lobe patients with asymmetrical patterns and their individual memory performance.*

tient #111 were completely void of neurons. These patients with sevére hippocampal sclerosis performed as poorly preoperatively as they did following removal of the entire anterior temporal lobe.

At this point, we can make the following conclusions regarding the performance on the delayed-recall of unrelated word-pairs and the left (dominant) hippocampus:

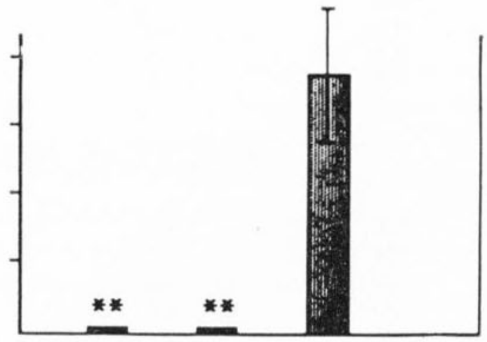
1. Minimal hippocampal damage does not impair performance.
2. Extensive damage limited primarily to either the anterior or posterior section of the hippocampus is not sufficient to severely impair performance.
3. Extensive damage in both the anterior and posterior sections is correlated with severely impaired performance.

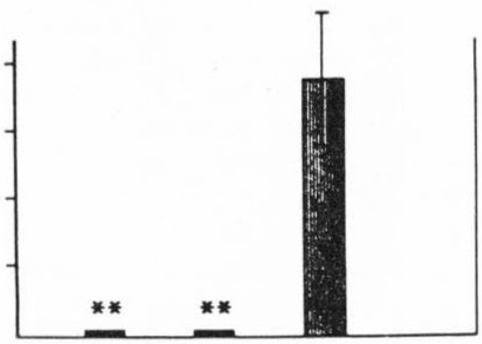
PATIENTS WITH EXTENSIVE DAMAGE EXTENDING ANTERIORLY AND POSTERIORLY

MEMORY SCORES

PRE-OP POST-OP N.C.

PATIENT #	ANT	POST
111		
FDU	71	69
FDL	76	63
CA4	100	90
CA3	72	90
CA2	100	75
CA1	97	100
PRO	85	100
SUB	18	37
PRE	0	0
204		
FDU	67	62
FDL	79	53
CA4	87	90
CA3	86	86
CA2	-	-
CA1	93	93
PRO	74	96
SUB	1	33
PRE	0	26





** > 2 SD below normal controls

Fig. 7. — *Neuron loss (expressed as percent reduction from normal) of two left temporal lobe epileptics with extensive hippocampal damage and their individual memory performance.*

PATIENTS WITH SELECTIVE DAMAGE EXTENDING ANTERIORLY <u>AND</u> POSTERIORLY

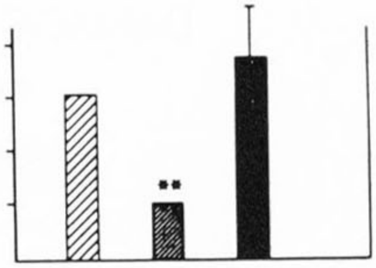
ANT POST — AT LEAST CA1 + PRO DAMAGE

MEMORY SCORES — PRE-OP POST-OP N.C.

PATIENT #

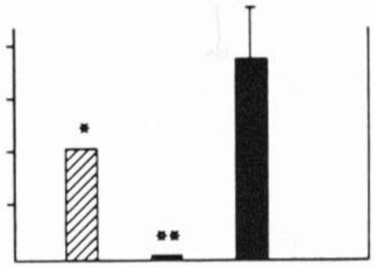
G3

	ANT	POST
FDU	60	45
FDL	79	55
CA4	30	65
CA3	44	76
CA2	68	72
CA1	90	87
PRO	100	96
SUB	12	84
PRE	0	0



133

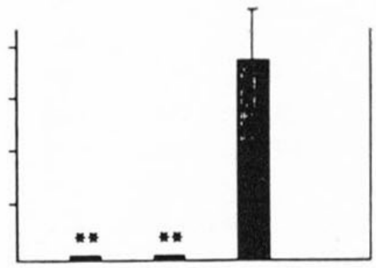
	ANT	POST
FDU	-	62
FDL	-	53
CA4	-	95
CA3	-	83
CA2	-	56
CA1	93	97
PRO	93	100
SUB	18	80
PRE	0	35



ONLY CA1 DAMAGE

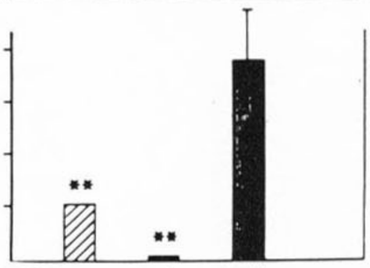
175

	ANT	POST
FDU	64	69
FDL	73	66
CA4	49	75
CA3	65	79
CA2	54	53
CA1	97	93
PRO	48	13
SUB	0	14
PRE	17	25



210

	ANT	POST
FDU	[illegible]	68
FDL	76	52
CA4	62	45
CA3	30	48
CA2	46	59
CA1	90	87
PRO	40	29
SUB	0	0
PRE	0	0



* > 1 SD below normal controls

** > 2 SD below normal controls

Fig. 8. — *Neuron loss (expressed as percent reduction from normal) of four patients with restrictive cell loss and their individual memory performance.*

PATIENTS WITH SELECTIVE HIPPOCAMPAL DAMAGE:

Patients with marked damage restricted to selective areas of the hippocampus provided an opportunity to address the functional importance of specific subregions. Four patients with marked damage to selective regions which extended anteriorly and posteriorly are shown in Figure 8.

There are data from two subgroups of patients shown in Figure 8: a) two patients with at least marked damage to the CA1 and the adjacent prosubicum extending throughout the anterior and posterior sections (Patients #G3 and #133) and b) two patients with damage restricted to the CA1, also extending throughout the hippocampus (Patients #175 and #210). There were contrasting memory results; the two patients with marked CA1 and prosubiculum damage (87 to 100 % cell loss in these areas) performed reasonably well on the word-pairs task before surgery. In contrast, the two patients with only CA1 damage performed poorly before and after surgery.

One possible explanation for this variability is that these anatomical areas are not critical for intact performance on the delayed-recall of unrelated word-pairs. If the anatomical areas are markedly damaged and the patients can perform the behavior, then it is reasonable to assume that these areas are not necessary for that task.

Another explanation for the variable performance of these selectively-damaged patients, one which appears to be more compatable with the recent report of Zola-Morgan et al. (1986), may lie in the time course of their hippocampal lesions. All four patients in this group had at least one seizure before the age of 12 years. The cell loss probably represents longstanding damage which would allow compensatory mechanisms, such as axonal sprouting and synaptic reorganization (cf., Cotman and Lynch, 1976 and Schneider and Jhavari, 1974) by the less-affected subregions. Those patients described earlier with extensive damage throughout many subregions of the hippocampus would not have the potential for such compensatory reorganization. Since we documented left hemisphere language dominance by the intracarotid Sodium Amytal procedure, the contralateral right hippocampus did not appear to underlie this behavior. Moreover, we showed the dependency of this measure on the left temporal lobe by impaired performance after left temporal lobe surgery.

DISCUSSION

These studies on the memory deficits in temporal lobe epileptics provide information relevant to the anatomical substrate of memory. We have identified a behavioral measure, delayed-recall of unrelated wordpairs, which is dependent upon the integrity of the left hippocampus. Furthermore, we have shown the extent of hippocampal damage necessary to disrupt performance on this task. The behavior is consistently disrupted by extensive neuron loss (> 80 % reduction from normal) that extends throughout the anterior-posterior axis of the hippocampus. Extensive damage limited to either the anterior or posterior section is not sufficient to consistently impair performance. The resiliency of the hippocampus to function with less than widespread damage has been implicated by two other findings. First, the efficiency of the ISA procedure which pharmacologically aborts one hemisphere, while testing the memory capability of the contralateral hemisphere, is reduced if the ipsilateral posterior hippocampus is not perfused. We have found that memory competence may be maintained during ISA injection of the hemisphere contralateral to severe hippocampal damage if the ipsilateral posterior cerebral artery is not perfused by the injection (Rausch et al., 1988). These results imply that the unperfused posterior hippocampus of the injected hemisphere may be maintaining memory functions. Second, we reported on an unfortunate case of a woman who underwent an amygdalohippocampectomy and subsequently became amnestic (Rausch et al., 1985). Pathological analyses of the removed hippocampus indicated that the patient had severe hippocampal damage posteriorly, but several hippocampal regions intact in the anterior section. Apparently, prior to surgery, the relatively intact anterior hippocampal regions were capable of maintaining general memory functioning.

Yet, in spite of the resiliency of the remaining neurons in the hippocampus to maintain functioning, the hippocampal tissue is tightly committed to its associated behavior. With extensive hippocampal damage, the correlative behavior is consistently and severely impaired. There does not appear to be recovery from the deficit. This occurs with both unilateral resections, as in the studies described above, as well as bilateral resections. In regards to the latter, bilateral temporal lobe resections are known to result in a permanent, unrelenting, severe amnestic syndrome (Scoville and Milner, 1957). Interestingly, the behavioral correlates of newer cortical regions, such as the hippocampal gyrus and lateral neocortex, do not appear to be as tightly associated with their behavioral correlates (Rausch and Babb, 1987; 1988). Damage in the neocortical areas significantly impairs the behavior of groups of patients, but not as consistently or as severely as seen with behavioral deficits associated with hippocampal lesions.

A potential role of the amygdala in the present findings cannot be ruled out. However, there are several indications that this role is minimal. The amygdala is by anatomy closely associated with the anterior hippocampus. Thus, the integrity of the amygdala may be best estimated from that of the anterior hippocampus. Yet, we have found that anterior hippocampal damage does not correlate with our behavioral measure; a behavioral deficit requires damage throughout the hippocampus. In addition, the two patients in these analyses who were noted at surgery to have "toughness" of the amygdala performed very well on the delayed-recall of the unrelated word-pairs.

Additional research is underway to further delineate the memory process associated with the hippocampus. The potential contributions of other temporal lobe structures to these cognitive processes are being investigated by studying patients with atypical pathology and diverse surgical treatments.

SUMMARY

Temporal lobe epileptics frequently have as their pathological substrate selective neuron loss of the hippocampus. The extent and pattern of the hippocampal cell loss vary among patients. These patients also exhibit varying degrees of memory loss. We have demonstrated a relationship between the memory loss in left temporal lobe epileptics and their hippocampal lesions. Specifically, we have identified a memory measure, delayed-recall of unrelated word-pairs, that is highly dependent upon integrity of left hippocampus. We have also shown that extensive damage throughout the hippocampus is necessary to consistently impair this measure. These findings are relevant to understanding the memory loss of temporal lobe epileptics as well as providing insight into our understanding of the anatomical basis of memory.

RESUMEN

Epilépticos de lóbulo temporal frecuentemente tienen como substrato patológico, una pérdida neuronal selectiva al nivel del hipocampo. La extensión y la modalidad de la neurodegeneración hipocámpica varía en los distintos pacientes. Estos pacientes presentan también grados diferentes de pérdida de memoria. Hemos demostrado en epilépticos del lóbulo temporal izquierdo una relación entre la pérdida de memoria y sus lesiones hipocámpicas. Más específicamente hemos puesto a punto una medida de la memoria, un recuerdo en el tiempo de grupos de palabras no relacionadas entre ellas lo cual depende principalmente de la integridad del hipocampo izquierdo. Nosotros hemos igualmente mostrado que una lesión propagada a todo el hipocampo es necesaria para modificar de modo significativo esta medida.

Estos resultados contribuyen a la comprensión de la pérdida de memoria en los epilépticos del lóbulo temporal así que a nuestra comprensión del soporte anatómico de la memoria.

RÉSUMÉ

Il est fréquent chez des épileptiques du lobe temporal d'observer comme substrat pathologique une perte neuronale sélective au niveau de l'hippocampe. L'étendue et le schèma de neurodégénérescence hippocampique varie selon les patients. Ces patients présentent également divers degrès de perte de mémoire. Nous avons démontré chez des épileptiques du lobe temporal gauche une relation entre la perte de mémoire et leurs lésions hippocampiques. Plus spécifiquement, nous avons mis au point une mesure de la mémoire, un rappel dans le temps de paires de mots sans relation, qui dépend principalement de l'intégrité de l'hippocampe gauche. Nous avons également montré qu'une lésion propagée à tout l'hippocampe est necessaire pour modifier de manière significative cette mesure. Ces résultats contribuent a la compréhension de la perte de mémoire chez des épileptiques du lobe temporal ainsi qu' à notre comprèhension du support anatomique de la mémoire.

ZUSAMMENFASSUNG

Patienten mit Temporallappenepilepsie haben als pathologisches Substrat oft einen selektiven Verlust von Neuronen im Hippocampus. Das Ausmaβ und die Verteilung dieses Zellverlustes variiert zwischen den Patienten. Zudem zeigen diese Patienten verschiedene Ausmaβe von Gedächtnisstörungen. Wir konnten eine Beziehung zwischen Gadächtnisstörungen und Läsionen im Hippocampus bei Patienten mit Epilepsien, die im linken Temporallappen beginnen, zeigen. Im besonderen wurde ein Gedächtnisparameter - Wiederholung beziehungsloser Wortpaare nach einer Latenzzeit identifiziert, der in höchstem Maβ von der Integrität des linken Hippocampus abhängig ist. Weiters konnte gezeigt werden, daβ eine beträchtliche Zerstörung des gesamten Hippocampus notwedig its, um dieses Gedächtnismaβ konsistent zu beeinträchtigen. Diese Ergebnisse sind von Bedeutung für das Verständnis der Gedächtnisstörungen bei Patienten mit Temporallappenepilepsie sowie für die anatomische Basis des Gedächtnis im allgemeinen.

ACKNOWLEDGEMENTS

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Memory Traces: How to Increase and Decrease Their Strength

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Memory traces are physical entities that exist inside organisms. Most scientists would accept the idea that memory must in some way involve the functional connections of neurons. Today there is good evidence that circuits of neurons underlie simple conditioned responses in mammals (Thompson and Donnegan, 1986). Ramon y Cajal, an early thinker on the subject, suggested that learning was an adaptation of the brain and that perfection of learning through exercise could be attained in the central nervous system by "...the passage of the impulse or the formation of new cell processes ... capable of improving the suitability and the extension of the contacts, and even of making entirely new connections between neurons primitively independent" (Ramon y Cajal, 1937, p. 459). Thus Ramon y Cajal not only postulated that circuits of neurons were the substrate of memory formation; he also thought that changes in synaptic contact were the mechanism by which the brain stored information.

The concept of circuits of neurons forming the neural substrate of memory was clearly elaborated by Hebb (1949) in his postulation of the existence of a *dual trace mechanism*. In this conception, memory was first encoded as a reverberating cellular assembly of neurons. The activity of this assembly would decrease over time unless exercised by repetition; this assembly formed the basis of short-term memory Through exercise this assembly would become a functionally intact system through the growth of synaptic knobs and would become long-term memory (Hebb, 1949, see p. 65). Hebb's notion is remarkably similar to that of Ramon y Cajal. Recent studies suggest that synaptogenesis may in fact be fundamental to some kinds of learning in higher mammals (Black and Greenough. 1986).

Let us consider a possible form of a cellular assembly. The simplest form of a memory circuit of which we can conceive is two neurons with reciprocal innervation, as shown in Figure 1a. A circuit such as this is not of much use unless it is connected to an effector, as depicted in Figure 1b. Let us make this circuit more complex by considering exercise, as did Ramon y Cajal and Hebb.

Exercise is a concept central to learning; Thorndike postulated the *Law of Exercise* to explain the notion that there was a strengthening of connections with practice (Hilgard, 1956). However, the memory circuit depicted in Figure 1b does not know to keep up reverberatory activity that likely terminates as short-term memory decays. There must also be an input from a sensory or other modality that regulates in some way the activity of the circuit and hence determines its strength. Thus we can con-

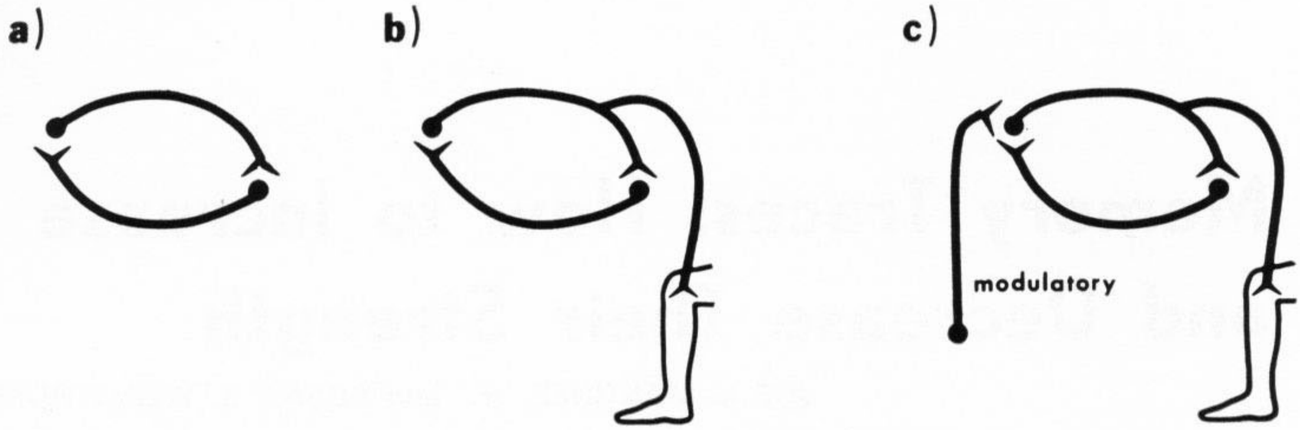

Fig. 1. — *a) A simple memory circuit: two neurons with reciprocal innervation.*
b) A simple memory circuit: connected to an effector.
c) A simple memory circuit: containing both an effector and a modulatory input to regulate the activity of the circuit.

ceive of a simple memory circuit, as shown in Figure 1c, with both an input and an output. Conceptualized in this way, we can see that the input is an integral part of the memory circuit. Furthermore, understanding how this input functions may provide the key to understanding a major aspect of associative processes in the CNS, since this input may regulate the strength of memory This paper is concerned with understanding this input to the memory trace.

Kety suggested in 1970 that there might be modulatory inputs to the memory trace; that is, an input might act to make memory trace; that is, an input might act to make memory traces stronger (see Figure 1c). Of course, if this modulatory input can make memory traces stronger, then it is possible that this input can make memory traces weaker as well (Martinez, 1986). The focus of this paper will be to describe conditions under which memory traces can be made stronger or weaker, and to consider what we know about these modulatory inputs, including their location(s) and sensitivity to pharmacological treatment.

The importance of a modulatory input can be appreciated by considering what Brown and Kulik (1977) called *flashbulb memories.* Normally, as we all know, memory is painfully engraved into the substrate of our brain through exercise. Yet sometimes events that happen only once in our lifetimes are remembered with extraordinary fidelity. These events may be uniquely individual, such as witnessing the birth of a child, or they may occur for an entire population, such as occurred when President John Kennedy was assassinated. Ask people who were alive at this to recount this event, and you will probably be surprised at the richness of detail of memory that surrounds this event in each person. The modulatory input to the memory trace may act to mark the salience of such events for the individual organism (Gold and McGaugh, 1977, Leshner, Merkle, and Mixon, 1981; Martinez, 1986). Sufficient activation of the modulatory system may allow memories to be stored permanently with a single exposure or trial.

In animal research, the importance of the event is communicated to an animal by the strength of the unconditioned stimulus. Interestingly, associative strength, as measured by retention performance, is not a simple increasing function of UCS intensity. Yerkes and Dodson (1908) showed almost 80 years ago that avoidance performance of the "dancing mouse" was a U-shaped function of footshock intensity; the best performance was observed with intermediate shock intensities. This observation of U-shaped functions is one to which we will return shortly.

Consider now the living situation of the average laboratory rodent-in a steel cage, in a climatically-controlled room, with other members of its species. Most likely this is not an especially stimulating environment.

One day a scientist many times the rodent's size comes along, takes the rodent to a laboratory, and places it into a conditioning situation. All of a sudden lights and buzzers start going on and off, bars may come flying out of walls of the chamber, or mild shock may suddenly come out of the floor. We believe this is probably a salient event in the life of the rodent, akin to those situations that produce flashbulb memories in humans. In fact, enduring one-trial learning may be observed in rats trained in either a passive avoidance or a conditioned taste aversion paradigm (Martinez and Rigter, 1983). In neurobiological terms, we would expect this type of conditioning situation to produce a high level of activation of the modulatory input to the memory trace. One research strategy for investigating this modulatory input is to administer pharmacological agents with specific actions on a known neurobiological system, and to observe any resultant changes in retention performance. Many such pharmacological agents and other treatments have been studied using this strategy and are known to either enhance or impair retention performance (Martinez, Jensen, and McGaugh, 1981).

How does one determine whether a particular treatment is modulatory, as opposed to having an action on another element of the memory trace? A good example of a modulatory treatment is provided by the research of Gold and van Buskirk (1976). In this study groups of rats were trained in a passive avoidance task at different footshock intensities, and then were given ACTH (3 or 6 IU) immediately following training. The rats were given a retention test 24 hours later. At the lowest footshock level used (0.4 mA), both doses of ACTH enhanced memory. At the next highest level (0.5 mA) only the lower ACTH dose was enhancing, and as the footshock was increased further (0.7 or 2.0 mA) both doses of ACTH produced retrograde amnesia. Thus, whether enhancement or impairment was observed depended on both the level of the training footshock and the strength of the experimental treatment. In other words, the ACTH treatment and the UCS interacted in such a way as to suggest that they were acting on a common mechanism as regards retention of the memory. Such a common mechanism, clearly capable of producing both enhancement and impairment, could easily be the modulatory input to the memory trace.

Another line of evidence suggesting that some pharmacological treatments act as modulators is the U-shaped dose-response curves they frequently produce. An example is seen in a study in which amphetamine was administered i.p. to rats immediately after training in a passive avoidance conditioning task. The rats were given a retention test 72 hours after training. An intermediate dose of amphetamine (1.0 mg/kg) was found to enhance retention performance, whereas both lower (0.25 mg/kg) and higher doses were ineffective (4.0 mg/kg) (Martinez, Jensen, Messing, Vazquez, Soumireu-Mourat, Geddes, Liang, and McGaugh, 1980). Recall that a U-shaped function also was observed by Yerkes and Dodson (1908) when they trained mice under increasing intensities of footshock. Thus, the fact that increasing the intensity of the UCS and increasing the dose of the experimental treatment both produce a similar U-shaped function suggests that the UCS and the pharmacological treatment act on a common mechanism to increase associative strength.

In agreement with the idea that the UCS and the experimental treatment act on a common mechanism is the important observation that the experimental treatment can be substituted for practice. Animals can be trained to a criterion level of performance in fewer trials if training is accompanied by an appropriate pharmacological treatment. For example, Rigter, Jensen, Martinez, Messing, Vazquez, Liang, and McGaugh (1980a) reported that D-Ala^2-[D-leu]enkephalin (DADLE) admininstered to rats before training in a multi-trial passive avoidance task resulted in the animals requiring fewer trials to reach a criterion of staying in the start compartment for 200 seconds. Thus a similar level of associative strength was achieved by either a larger number of training trials, or by a smaller number of training trials combined with an appropriate pharmacological treatment. Given that the UCS and the experimental treatment can substitute for one another, it is to be expected that the experimental treatment can act as a UCS. This was shown by Introini and Baratti (1984), who found that beta-endorphin given to mice following passive avoidance training in the absence of footshock produced significant retention when the animals were tested on the following day. Interestingly, the strength of memory on the test day was related to the dose of beta-endorphin the mice received on the previous day. It is reasonable to expect that, under certain conditions, activation of a modulatory system can produce strong learning in one trial, and under these conditions we may have an animal equivalent of a flashbulb memory.

Further evidence that practice and the experimental treatment act via a common mechanism to modulate the memory trace is provided by the observation that pharmacological treatment is only effective when administered concurrently with the training. For example, neither [leu]enkephalin (Dana and Martinez, 1986), DADLE (Rigter et al., 1980a), nor beta-endorphin (Introini and Baratti, 1984) alters avoidance responding in animals when the drug treatment is not given in close temporal proximity to the training experience. Further, once learning is established through practice, it is no longer susceptible to additional pharmacological modulation. Thus Mauk, Castellano, Rodeout, Madden, Barchas, and Thompson (1983) reported that overtraining protects the conditioned nictitating response of rabbits from disruption by opiates.

If one assumes that modulatory systems must exist, then it is appropriate to ask what we know about their relationship to the memory trace. The simplest conceptualization is to assume that a peripherally administered pharmacological agent passes directly into the brain to affect the memory trace. Such a conclusion was reached by de Wied (1964) in studying the effects on memory of ACTH, vasopressin, and analogs and fragments of these hormones. This model is depicted in Figure 2. The conclusion that these substances have a direct action on the CNS was based on the observation that they produce parallel effects on memory after central or peripheral administration, as well as on a comparison of the magnitude of the effective dose following administration by these routes (Bohus and de Wied, 1981; Martinez, 1986). With many drugs central administration produces effects on behavior at doses 10 to 100 times less than are effective following peripheral administration. However, there is little evidence that ACTH, vasopressin, or their fragments pass the blood-brain barrier and considerable evidence that they do not (Allen, Kendall, McGilvra, and Vancura, 1974; Lebrun, Le Moal, Koob, and Bloom, 1985; Pardridge, 1983).

A recent conception of how and where neuropeptides act to influence memory suggests that the primary site of action may be outside the blood-brain barrier, and that their modulatory effect may be mediated through autonomic afferents (Ettenberg, van der Kooy, Le Moal, Koob, and Bloom, 1983), as depicted in Figure 3. An alternative view is provided by Sahgal (1984), who suggests that modulatory substances may affect the memory trace only indirectly, through a primary action on arousal (or motivation or attention). This notion is presented in Figure 4.

Our own view of how the modulatory system operates is based on our work with one family of opioid peptides, the enkephalins. We suggest that [leu]enkephalin may directly affect memory traces by an action on the modulatory inputs to these circuits, even though [leu]enkephalin itself does not cross the blood-brain barrier in sufficient quantities to be of physiological significance (Martinez, Weinberger, and Schulteis, submitted). How this might occur is presented in Figure 5. In this view the modulatory substance ([leu]enkephalin]) acts either on autonomic afferents returning to the CNS or on circumventricular organs which, while part of the CNS, lie outside the blood-brain barrier.

Our view of the memory circuit is that within each organism its various components are distributed in nature. Thus if one

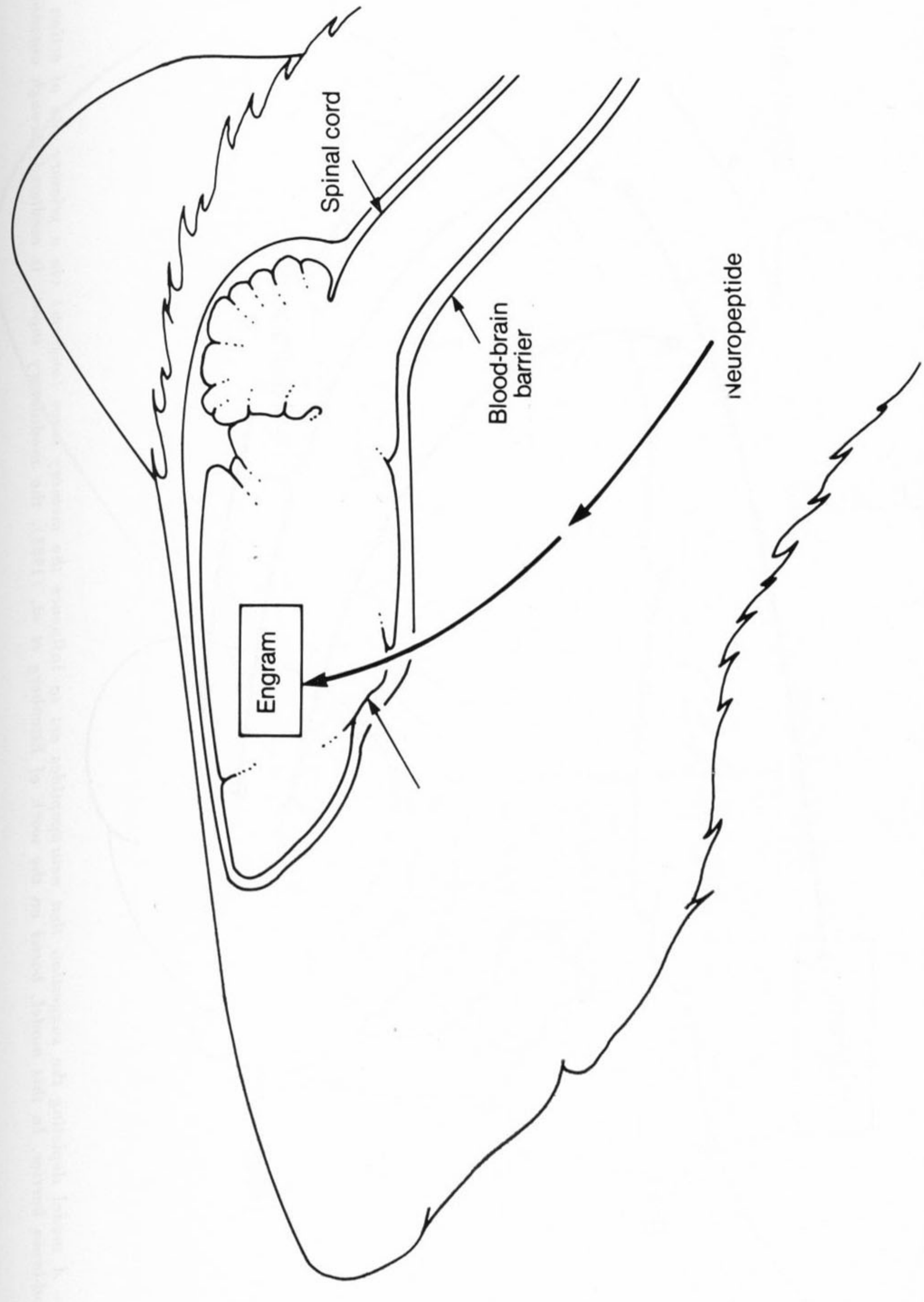

Fig. 2. — *A model of how neuropeptides modulate the memory trace (engram), based on de Wied (1964). In this conceptualization, it is assumed that a peripherally administered pharmacological agent passes directly into the brain to affect the memory trace.*

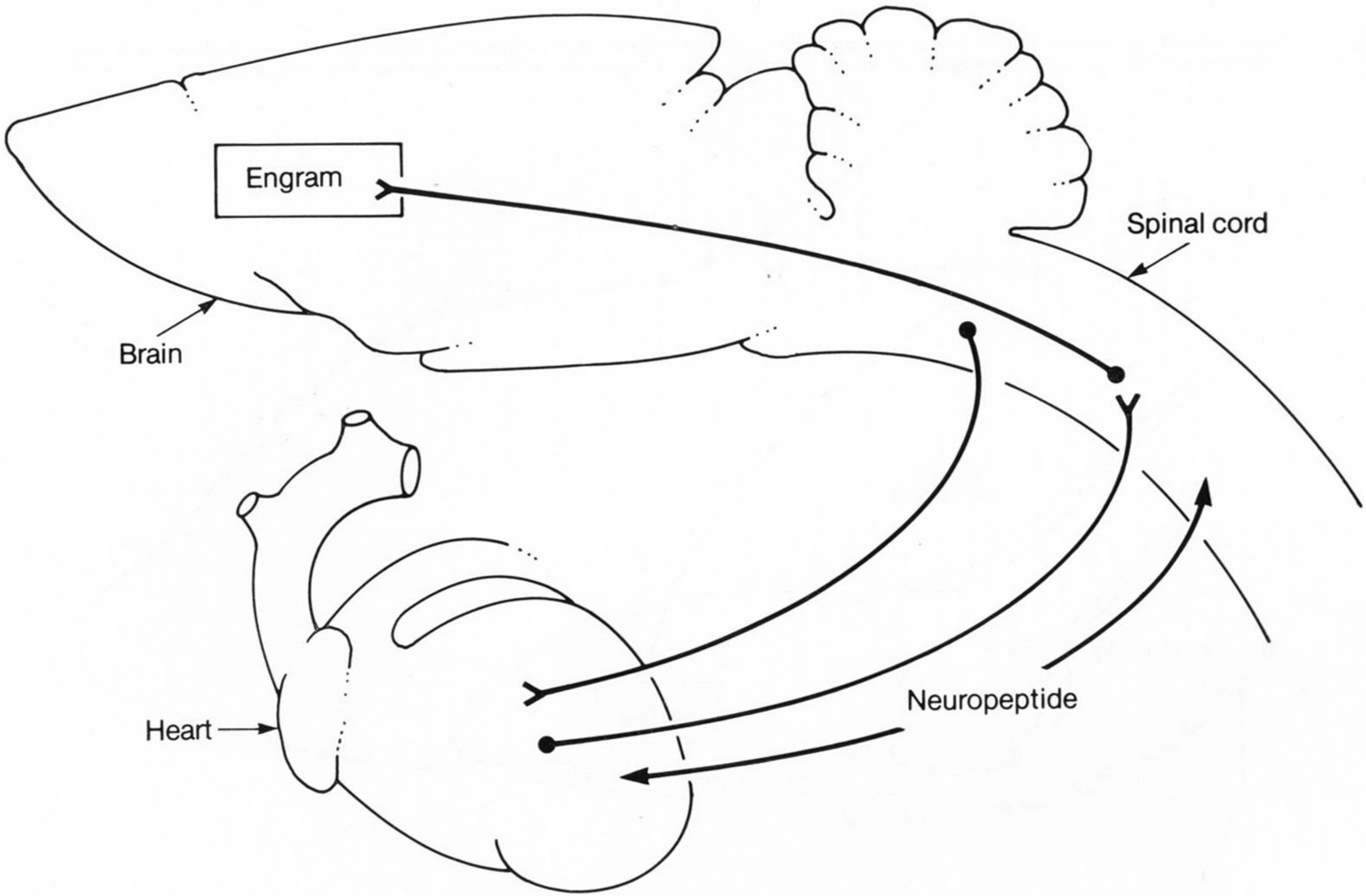

Fig. 3. — *A model depicting the suggestion that neuropeptides act to influence the memory trace (engram) via a primary site of action outside the blood-brain barrier. In this model, based on the work of Ettenberg et al. (1983), the modulatory input is mediated through autonomic afferents.*

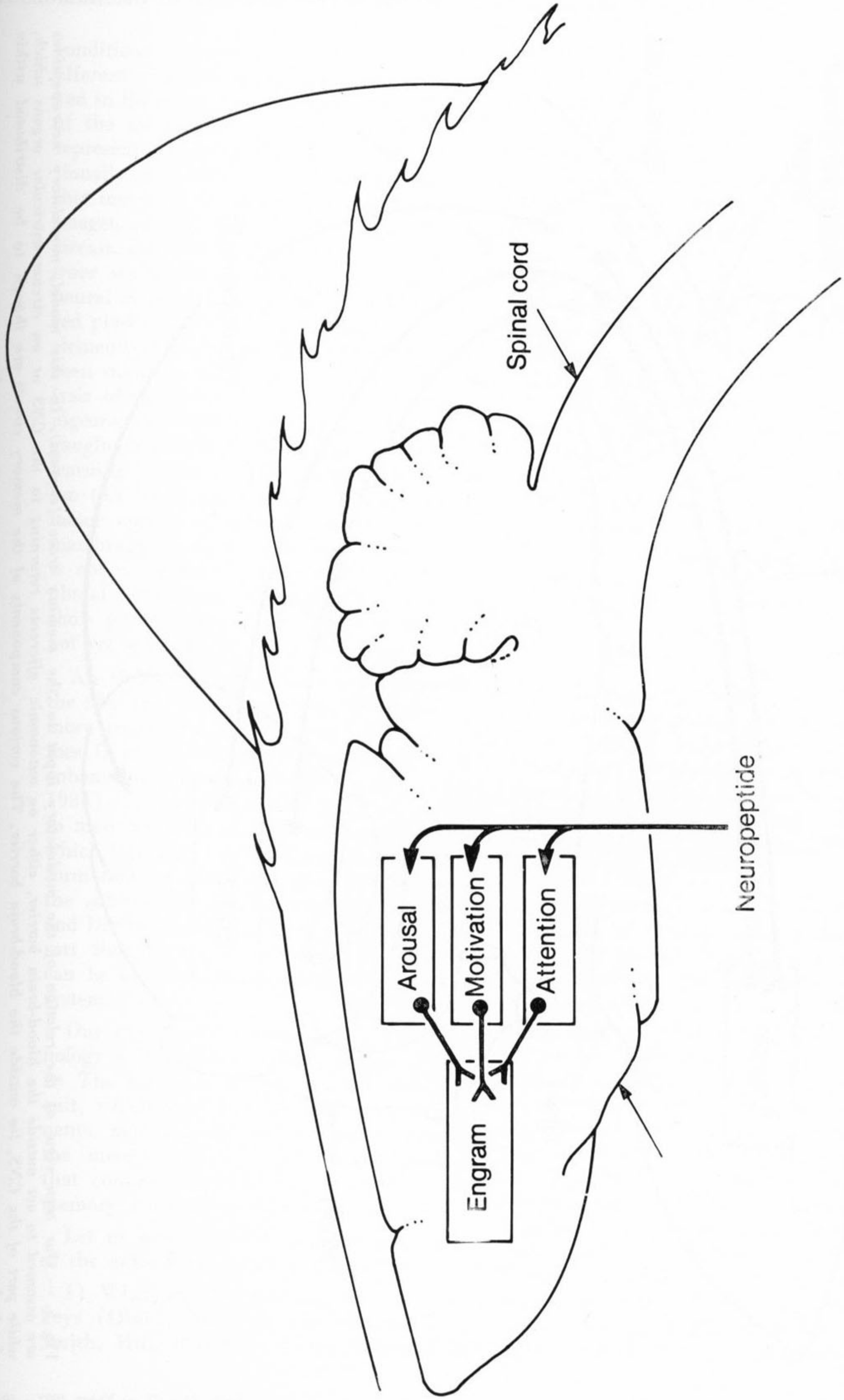

Fig. 4. — *A model depicting the suggestion of Sahgal (1984) that neuropeptides modulate the memory trace (engram) indirectly, through a primary action on arousal or motivation or attention.*

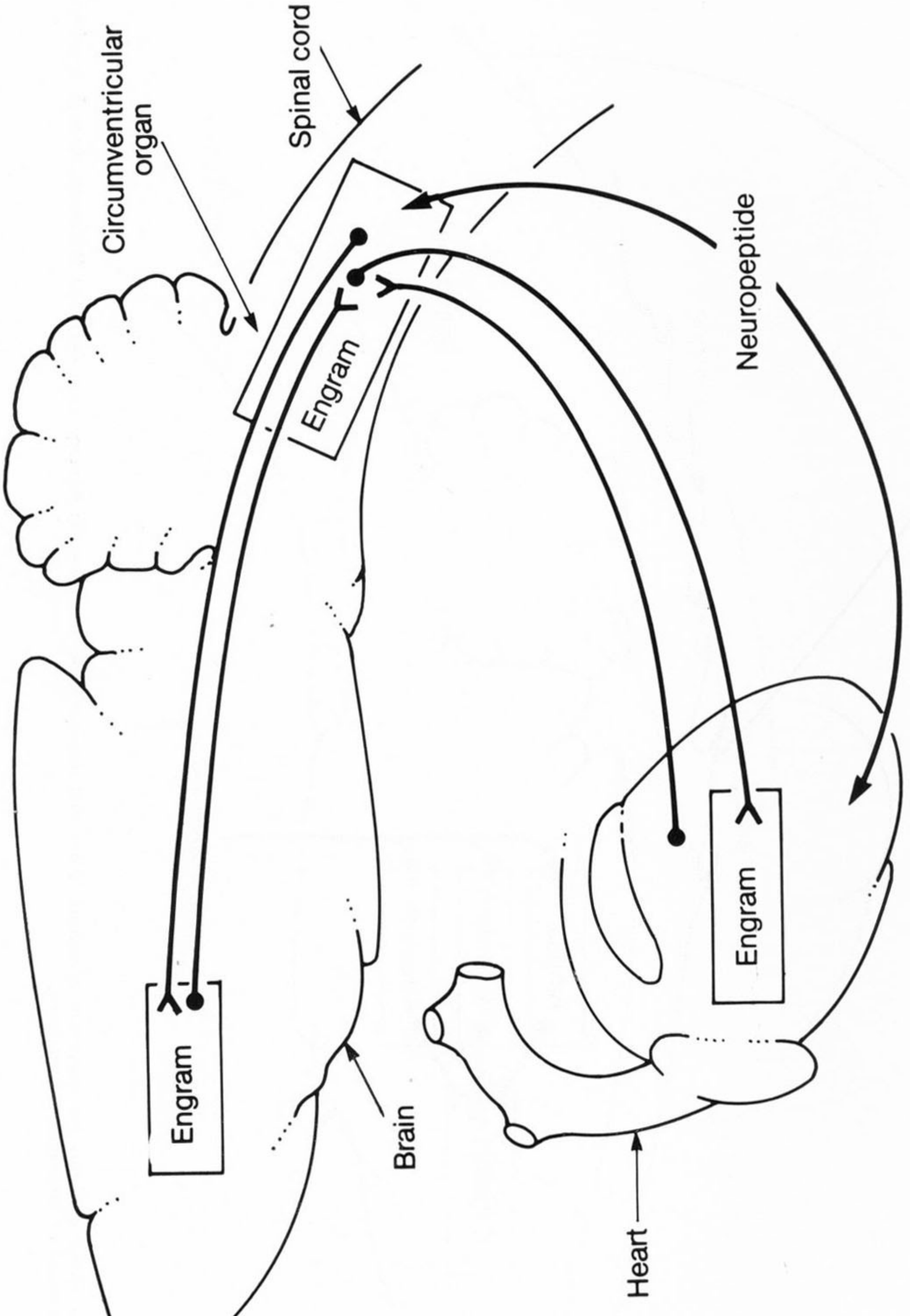

Fig. 5. — Our own view of the relation of the modulatory input to the memory trace (engram). In this model, modulatory neuropeptides are assumed to act outside the blood-brain barrier, either on autonomic afferents returning to the CNS or on circumventricular organs which, while part of the CNS, lie outside the blood-brain barrier. The various components of the memory circuit are though to be distributed within the body.

conditions a change in heart rate, all the afferent and efferent neural elements involved in these heart rate changes must be part of the memory circuit, since neurons that represent the memory trace must be functionally connected to autonomic efferents that innervate the heart. Thompson and Donnegan (1986) call this the *memory trace circuit,* and distinguish it from the *memory trace* which they define as the subset of neural elements that exhibit training-induced plasticity. Peripheral sensory and motor elements of memory trace circuits have not been shown to be plastic. For example, analysis of conditioned heart rate changes in pigeons shows that activity in the retinal ganglion does not change as a result of learning (Cohen, 1985). Similarly, Thompson (1976) found no plastic changes in the motor output of a conditioned nictitating membrane response in rabbits. However, it is not yet known with certainty that peripheral components of memory traces do not show plasticity in other forms of learning not yet studied.

An additional bit of evidence supporting the idea that peripheral components of memory trace circuits can show plasticity comes from the demonstration of long term enhancement in sympathetic ganglia (Libet, 1984). Long term enhancement is similar in many respects to long term potentiation, which is thought to be a model of memory formation in the CNS and possibly even the substrate of the memory trace (Teyler and DiScenna, 1986). Thus plasticity of the sort thought to be necessary for learning can be observed in the peripheral nervous system.

Our hypothesis, stated within the terminology of Thompson and Donnegan (1986), is: The elements of the memory trace circuit, which may include hormonal components, may provide a modulatory input to the memory trace. Further, the neurons that connect the modulatory input to the memory trace may show plasticity.

Let us now consider some characteristics of the enkephalin modulatory system:

1) When injected peripherally into monkeys (Olson, Olson, Kastin, Green, Roig-Smith, Hill, and Coy, 1979; Olson, Roig-Smith, Mauk, LaHoste, Coy, Hill, and Olson, 1981), rats (Kastin, Scollan, King, Schally, and Coy, 1976; Rigter et al., 1980a; Rigter, Hannan, Messing, Martinez, Vasquez, Jensen, Veliquette, and McGaugh, 1980b; Izquierdo and Dias, 1981; Martinez, Rigter, Jensen, Messing, Vasquez, and McGaugh, 1981), and mice (Martinez, Olson, and Hilston, 1984), enkephalins influence acquisition or retention of several different conditioned responses. For example, [leu] enkephalin, at doses of 30 and 100 µg/kg administered i.p. to mice 2 minutes before training in a one-way active avoidance task, impairs acquisition of the response (Figure 6). Notice that the dose-response function is U-shaped (Schulteis et al, in press). As noted above, enkephalins are effective at altering conditioning, when injected systemically, in quantities that most likely do not cross the blood-brain barrier (Cornford, Braun, Crane, and Oldendorf, 1978; Rapoport, Klee, Pettigrew, and Ohno, 1980; Pardridge, 1983; Zlokovic, Begley, and Chain-Eliash, 1985).

2) Enkephalin effects on conditioning are sensitive to integrity of the adrenal medulla. Adrenal medullectomy abolishes the impairment of avoidance conditioning produced by systemic administration of enkephalin (Izquierdo, Carrasco, Dias, Perry, Netto, Wofchuk, and Souza, 1982; Martinez and Rigter, 1982).

3) Preventing access of endogenous enkephalins to their peripheral receptors produces effects on conditioning opposite to those observed with enkephalin itself. This has been shown in two ways-either by passive immunization with a [leu]enkephalin antiserum (Martinez, de Graaf, Chavkin, and Dana, 1985a) or by receptor antagonism using methylnaloxonium, a quaternary form of naloxone which does not cross the blood-brain barrier (Martinez and de Graaf, 1985). An example of a receptor antagonism study is shown in Figure 7. In this study mice were injected systemically with methylnaloxonium 5 minutes before being trained in a discriminated Y-maze avoidance task. Mice given methylnaloxonium (1.0 mg/kg) made fewer errors than saline-treated animals, demonstrating en-

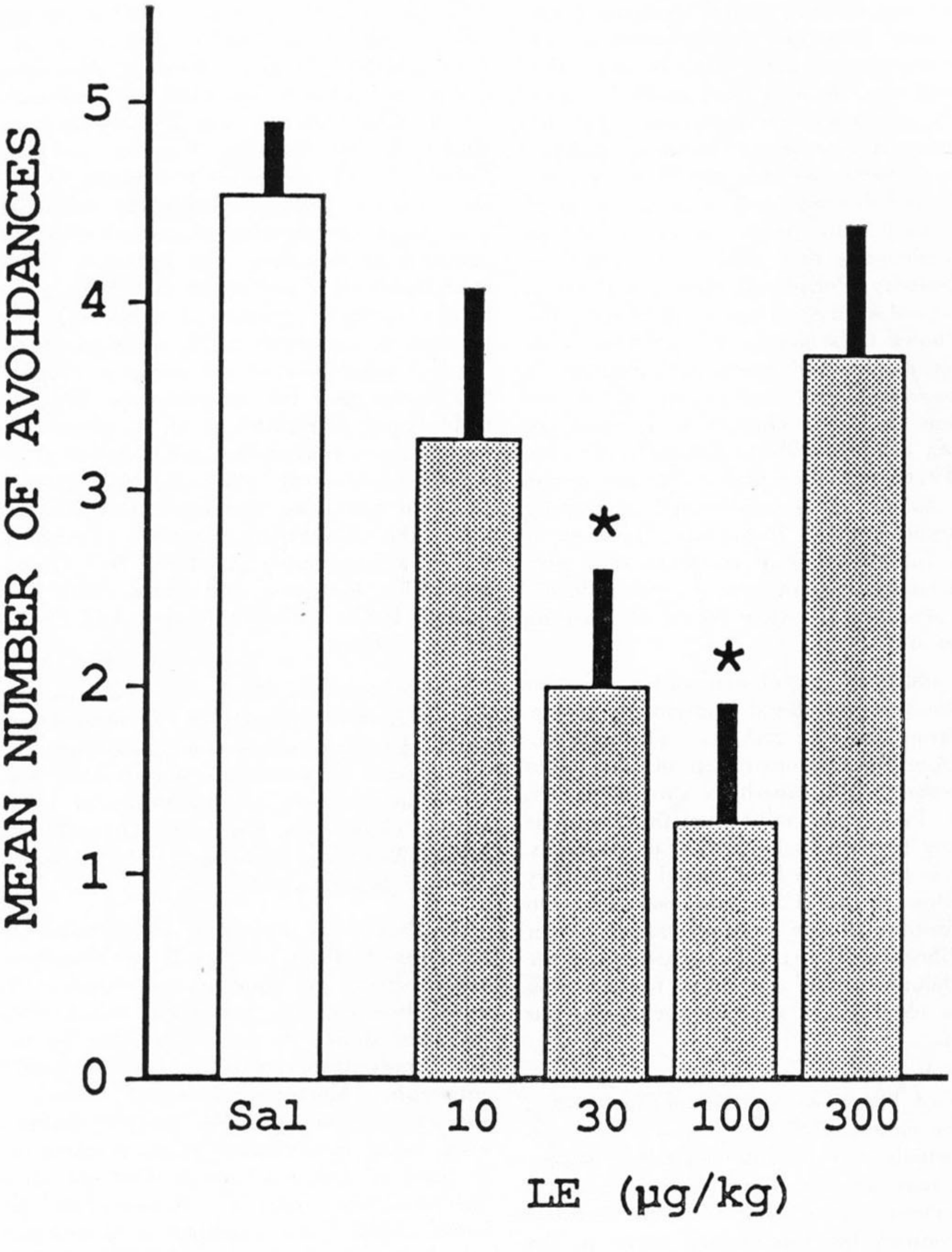

*Fig. 6. — [Leu]enkephalin (LE) impairs acquisition in mice of a one-way active avoidance response at odses of 30 and 100, but not 10 or 300, µg/kg. Drugs were administered 2 minutes before training began. Ordinate represents mean number of avoidances, expressed as % of the saline mean, ± S.E.M. over 14 training trials. * $p < 0.005$ versus control (saline) group. ** $p < 0.0001$ versus control (saline) group. Saline group, $N = 24$; all LE groups, $N = 10$ or 11. From Schulteis et al., in press.*

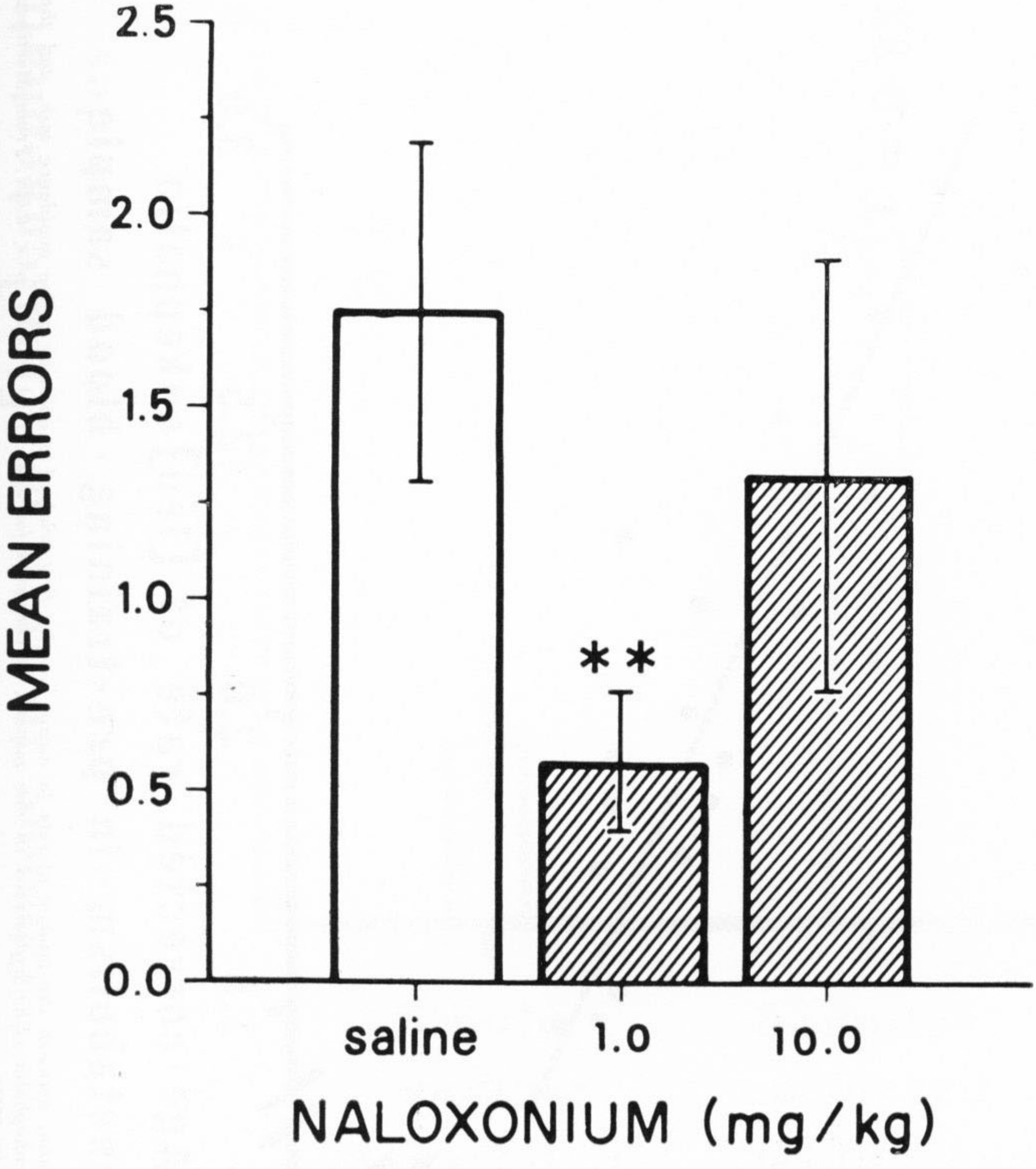

Fig. 7. — *Methylnaloxonium (naloxonium) enhances acquisition in mice of a discriminated Y-maze avodance task at a dose of 1.0, but not 10.0, μg/kg. Drugs were administered 5 minutes before training began. Ordinate represents mean number of errors ± S.E.M. over 8 training trials. ** $p < 0.05$ versus control (saline) group. All N = 9 — 12. From Martinez and de Graaf, 1985.*

hanced acquisition of the response (Martinez and de Graaf, 1985). As was seen with the learning impairment produced by [leu] enkephalin, the dose-response function for the learning enhancement produced by methylnaloxoniu mis also U-shaped. Importantly, the effects of both [met]- and [leu]enkephalin on avoidance conditioning can be antagonized by quaternary antagonists (Introini, McGaugh, and Baratti, 1985; Martinez and de Graaf, 1985), further supporting the interpretation that these neuropeptides have a primary site of action outside the blood-brain barrier. Together these results suggest that one major way in whcih memory traces may be made stronger or

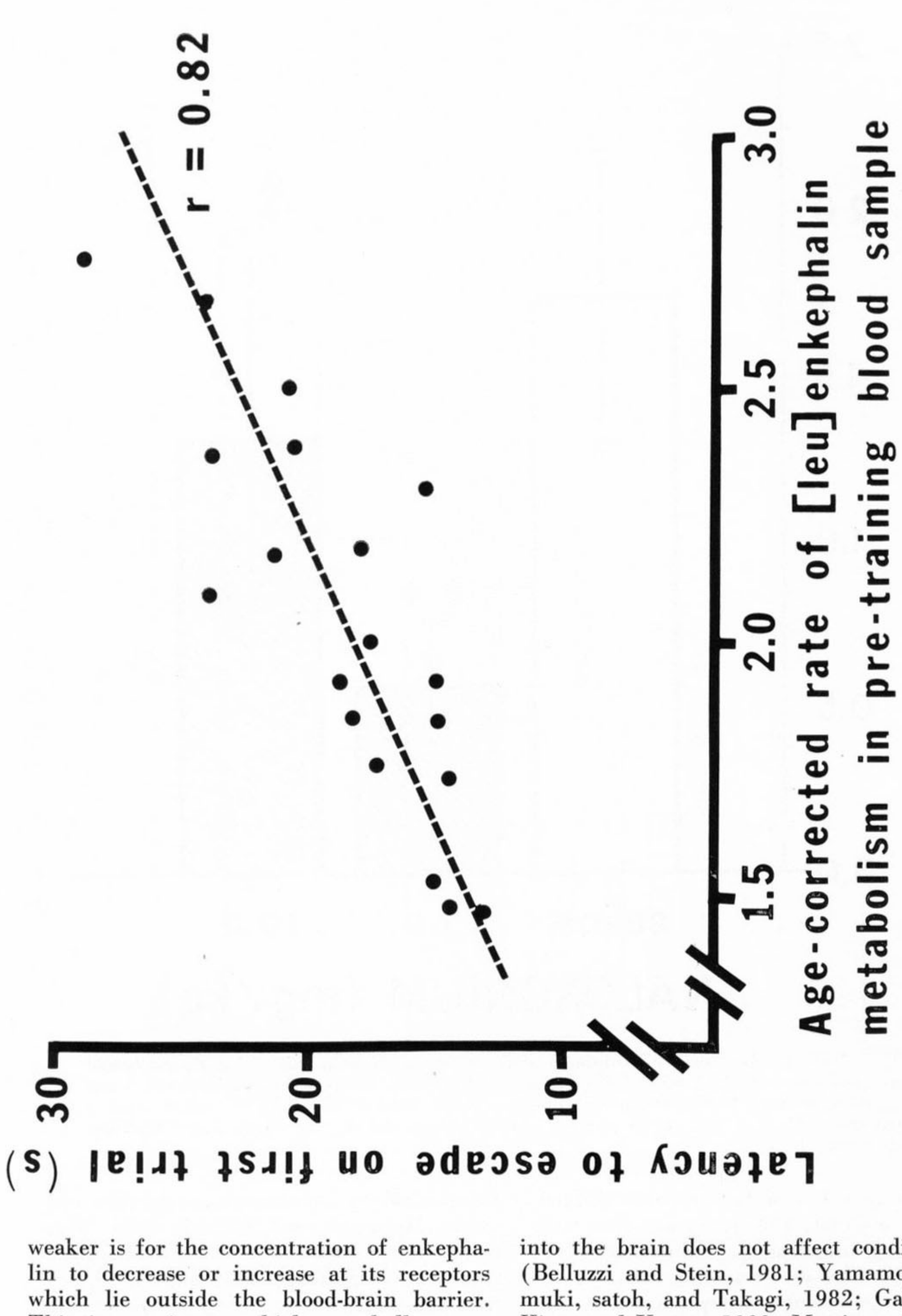

Fig. 8. — Correlation between the latency of rats to escape on the first trial of a one-way active avoidance task, and the amount of [leu]enkephalin (LE) hydrolyzed in one minute, as corrected for age: $r(19) = 0.82$, $p < 0.001$. From Martinez and Weinberger, in press.

weaker is for the concentration of enkephalin to decrease or increase at its receptors which lie outside the blood-brain barrier. This is a point to which we shall return shortly.

4) Infusion of [leu]enkephalin directly into the brain does not affect conditioning (Belluzzi and Stein, 1981; Yamamoto, Kamuki, satoh, and Takagi, 1982; Gallagher, King, and Young, 1983; Martinez, Conner, and Dana, 1985b). However, the opposite appears to be true for [met]enkephalin

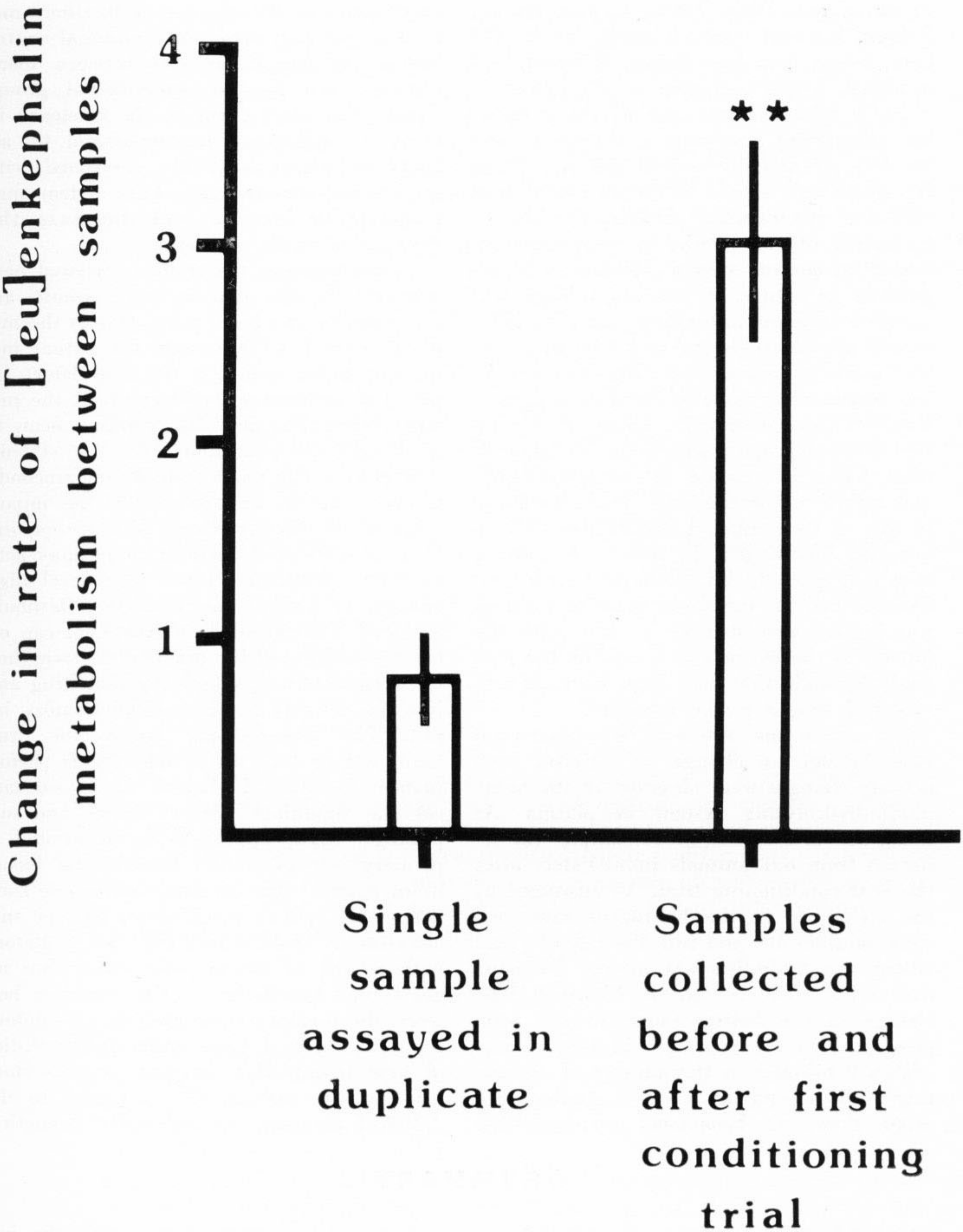

Fig. 9. — *Changes in percent of [leu]enkephalin (LE) metabolized between blood samples (mean absolute value of the difference between samples, as corrected for age by analysis of covariance, ± S.E.M.). Control group: difference in enzyme activity between sample pairs when a single sample was divided in half for duplicate assay; N = 12. Experimental group: difference in enzyme activity in blood samples collected immediately before and after the first conditioning trial; N = 18. ** p < 0.003 versus control group. From Martinez and Weinberger, in press.*

(Belluzzi and Stein, 1981; Lucion, Rosito, Sapper, Palmini, and Izquierdo, 1982; Fekete, Drago, Van Ree, Bohus, Wiegant, and de Wied, 1983; Gallagher et al., 1983).

5) A fifth characteristic of the peripheral enkephalin modulatory system is one we only recently discovered, and one which fits quite well with a hormonal modulatory role for enkephalins. Characterization of hydrolysis of enkephalin in rat plasma revealed an enzyme system which may be regulatory in nature. In this study blood was sampled through indwelling cannulae before and after a single training trial in a one-way active avoidance task. Enzyme activity was measured *in vitro* by incubating plasma with ^{3}H-[leu]enkephalin. Using a thin layer chromatography separation, metabolism rates were determined by comparing the amount of unmetabolized [leu]enkephalin to that of its combined metabolites (Martinez and Weinberger, in press). As can be seen in Figure 8, the results were startling. Enzyme activity measured prior to training was highly correlated ($r = 82$) with the latency of the animals to escape on the first trial. Animals that had high enzyme activity had longer escape latencies.

For an enzyme system to be regulatory it must be able to change its activity; such activity changes were observed in the enkephalin-hydrolyzing system in plasma. As noted above, a second blood sample was collected from our animals immediately after the first conditioning trial. As measured by the difference in the hydrolysis rates between samples one and two, the rate of [leu] enkephalin hydrólisis was altered following this trial, as can be seen in Figure 9. The change in metabolism rates between samples represents a potential 11.4 % change within 2 minutes in the amount of circulating [leu]enkephalin. Control studies indicated that the behavioral manipulations themselves, in the absence of footshock-motivated training, made only minimal contributions to the differences between blood samples seen in the experimental group. Thus, these data point to the existence in blood of a regulatory enzyme system, the activity of which is highly correlated with acquisition performance. This system may modulate or increase **and decrease** the strength of memory traces.

Taken together the studies reviewed here support the idea that memory circuits are composed of two basic parts. One is the memory trace that represents the actual memory and the second is the modulatory input that influences the strength of the memory trace. The modulatory input seems to be intimately associated with the systems that convey the magnitude of the unconditioned stimulus or presumably the importance of the present event. A single exposure to a sufficiently salient event may activate the modulatory input to a level high enough to produce an enduring flashbulb memory. Pharmacological treatment can often be substituted for practice to strengthen the memory trace, suggesting that drug and practice activate a common modulatory input. The dose-response curves for drug treatment or UCS amplitude versus performance are often U-shaped. Many substances may modulate memory traces, and surprisingly many appear to do so through a primary site of action outside the blood brain barrier. Our investigation of one such peripheral system revealed an enzyme mechanism in blood which may be regulatory with regard to escape peformance. We suggest that knowledge of the interface between the memory trace and its modulatory input may reveal some basic characteristics of how mammalian nervous systems store information, particularly in regard to the "glue of memory" or associative strength.

SUMMARY

In addition to the memory trace itself, the memory circuit may include a modulatory input that influences the strength of the memory trace. This modulatory input, which can make memory traces stronger as well as weaker, appears to be intimately associated with systems that convey the importance of the present event. A single exposure to a sufficiently salient event may activate the modulatory input to a level high enough to produce an enduring memory. Experimental investigation of this modula-

tory input indicates that pharmacological treatment can often be substituted for practice to strengthen the memory trace, suggesting that drugs or practice activate a common mechanism. Similarly, drugs and the UCS interact in such a way as to suggest they also act on a common mechanism to modulate retention. Dose-response curves for drug treatment or UCS amplitude versus performance are often U-shaped. Many drugs that modulate memory appear to do so through a primary site of action outside the blood-brain barrier. Our investigation of one such peripheral system, namely that for [leu]enkephalin, revealed an enzyme mechanism in plasma which may be regulatory with regard to escape performance. We suggest that investigation of the interface between the memory trace and its modulatory input may lead to understanding of how mammalian systems store information, particularly in regard to associative strength.

RESUMEN

El circuito mnésico contiene además del trazo mnésico un influjo modulador que ejerce una influencia sobre la energía del trazo mnésico. Este influjo modulatorio, que puede convertir los trazos en más fuertes o en más débiles, parece estar intimamente asociado a los sistemas que comunican la importancia del hecho actual. Una sola exposición a un hecho suficientemente notorio puede activar el influjo modulatorio a un nivel bastante alto para originar una memoria duradera. Las investigaciones experimentales muestran que los tratamientos farmacológicos pueden ser sustituídas por la repetición en el aumento del trazo mnésico; lo que parece indicar que las drogas y la repetición activan un mecanismo común. También la acción recíproca de los agentes farmacológicos y de los estímulos incondicionales sugiere que ellos actúan también sobre un mecanismo común para modular la retención. Las curvas que señalan las relaciones entre dosages y efectos de las drogas o las relaciones entre la fuerza del estímulo incondicional y el comportamiento, son a menudo en forma de U (curvas en forma de U). Muchas drogas que modulan la memoria parecen actuar sobre asientos fuera de la barrera hemato encefálica.

Nuestras investigaciones sobre uno de estos sistemas más particularmente el que implica la (leu) encefalina, han revelado un mecanismo enzimático en el plasma que podría tener una acción reguladora sobre el comportamiento de fuga. Nosotros creemos que investigaciones sobr las relaciones recíprocas entre el trazo mnésico y el influjo modulatorio podrían conducir a la comprensión como los diferentes sistemas en los mamíferos guardan información particularmente frente a la fuerza asociativa.

RÉSUMÉ

Le circuit mnésique contient, en plus de la trace mnésique, un influx modulatoire qui exerce une influence sur la force de la trace mnésique. Cet influx modulatoire, qui peut rendre les traces mnésiques plus fortes ou plus faibles, semble être associé intimement aux systèmes qui communiquent l'importance du fait actuel. Une seule exposition à un fait suffisemment saillant peut activer l'influx modulatoire à un niveau assez élevé pour produire une mémoire durable. Les recherches expérimentales montrent que les traitements pharmacologiques peuvent être substitues à la répetition en augmentant la trace mnésique; ce qui semble indiquer que les drogues et la répetition agissent sur le même mécanisme. De même, l'action réciproque des agents pharmacologiques et des stimuli inconditionnels suggère qu'eux aussi agissent sur un mécanisme commun pour moduler la rétention. Les courbes qui rapportent les relations entre dosages et effets des drogues ou les relations entre la force du stimulus inconditionnel et le comportement sont souvent en forme d'U (Ushaped curves). Plusieurs drogues qui modulent la

mémoire semblent agir sur des sites en dehors de la barrière hémato-encéphalique. Nos recherches sur un de ces systèmes, plus particulierement celui qui implique la [leu] enképhaline, ont révélé un mécanisme enzymique dans le plasma qui pourrait avoir une action régulatrice sur le comportement de fuite. Nous croyons que des recherches sur les relations réciproques entre la trace mnésique et l'influx modulatoire pourraient aider à comprendre comment les différents systèmes chez les mammifères emmagasinent les informations, et surtout la force des associations.

ZUSAMMENFASSUNG

Der Gedächtnisschaltkreis (memory circuit) enthält neben der Gedächtnisspur auch noch einen modulierenden "Input", der die Gedächtnisspur sowohl verstärken als auch abschwächen kann. Dieser modulierende Input scheint mit Systemen, die Wichtigkeit des gerade gegenwärtigen Ereignisses weiterleiten, eng verknüpft zu sein. Eine einzige Darbietung eines genügend salienten Ereignisses kann den modulierenden Input so stark aktivieren, dass eine dauerhafte Erinnerung ensteht. Experimentelle Befunde, dass pharmakologische Behandlung den Übungseffekt, der zur Stärkung der Gedächtnisspur führt, oft ersetzen kann, deuten darauf hin, dass Drogen und Übung den gleichen Mechanismus anregen. Darüberhinaus legt der Interaktionseffekt von Drogen und Reizdarbietungen nahe, dass beide Stimuli an einem ihnen gemeinsamen Mechanismus, der das Behalten reguliert, ansetzen. Sowohl Dosis-Effektkurven als auch Reizamplituden-Leistungskurven haben häufig eine U-Form. Viele gedächtnisbeinflussende Drogen scheinen an einer Primäraktionsstelle zu wirken, die ausserhalb der Blut-Hirnschranke liegt. In unserer Untersuchung eines solchen peripheren Systems, desjenigen für [Leu]Enkephalin, weisen wir einen enzymatischen Mechanismus im Plasma nach, der wahrscheinlich das Fluchtverhalten reguliert. Weitere Untersuchungen der Schaltstellen zwischen der Gedächtnisspur und dem zugehörigen modulierenden Input könnten erheblich zu unserem Verständnis der für Saugetiere typischen Informations-speicherung, insbesondere der Assoziationsstärke, beitragen.

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The Irrelevance of Mammillary Body Lesions in the Causation of the Korsakoff Amnesic State

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This presentation will focus on a small but important aspect of the anatomy of memory, namely, on the significance of the mammillary body lesions in the Wernicke-Korsakoff syndrome. The conventional view, first stated by Gudden, in 1891, is that the mammillary body lesions are responsible for the characteristic memory defect (Korsakoff psychosis) associated with Wernicke encephalopathy. No doubt this view stems from the prominance and invariable presence of mammillary body lesions in Wernicke disease. In fact, there have been several reports of individual or small numbers of cases of Korsakoff psychosis, in which the pathologic changes were said to be confined to the mammillary bodies. Grunthal (1939) for example, described a 40 year old man with progressive dementia and memory disorder in whom a craniopharyngioma had destroyed the mammillary bodies; the thalamus and hypothalamus were said to be spared, although they do not appear to be normal in the one photomicrograph that was shown to illustrate this feature. The patient described by Remy (1942) was a chronic alcoholic who died at age 60 of carcinoma of the larynx, 19 years after the onset of Korsakoff psychosis; in brain sections, the thalamus appeared small, but histologically the diencephalic structures were said to be intact, except for the mammillary bodies, which were atrophic and gliotic. Delay and Brion (1954), Gruner (1956) and, most recently. Torvik, (1987) have made the same point. Torvik, in a postmortem series of 21 chronic (inactive) cases of Wernicke encephalopathy found three cases in which the pathologic changes were restricted to the mammillary bodies; each of these patients was said to have shown "decreased memory" during life.

The evidence presented in these reports in hardly compelling. Information about the patients' mental symptoms is remarkably sparse and was usually' obtained retrospectively, from a review of the patients' records. Whether the mental state in these patients was truly representative of Korsakoff psychosis is, therefore, far from certain. Also there is no way of judging the thoroughness with which the brain, and particularly the thalamus, was examined histologically. The changes in the medial thlamus (of patients with Korsakoff psychosis) are often subtle and restricted in extent, and it is not inconceivable that they were overlooked.

Our studies of the Wernicke-Korsakoff syndrome have led to quite a different conclusion, viz., that the diencephalic lesions giving rise to the memory defect are those of the medial thalamus (medial dorsal nucleus and possibly the pulvinar) and not those of the mammillary bodies. The evidence for this point of view has been fully developed in our monograph on the Wernicke-Korsakoff syndrome (Victor et al, 1971), and will be summarized here in its briefest form. It is based essentially on two sets of observations:

(1) The diencephalon was carefully studied (often in serial section) in 38 patients who had been examined repeatedly during life and satisfied fully the diagnostic criteria of Korsakoff psychosis; at autopsy, each of these patients, without exception, showed lesions of the medial part of the medial dorsal nucleus of the thalamus. and most of them showed lesions of the medial pulvinar as well. Contrariwise, in patients who during life showed only the symptoms and signs of Wernicke encephalopathy and failed to develop a memory defect, the thalamus was never involved.

(2) There were five patients in our autopsy series who showed the characteristic pathologic changes in the mammillary bodies but had shown no memory defect during life. In one of them, the presence of a memory defect could be excluded with certainty, for he was under close neurologic observation when he died suddenly as a result of a ruptured gallbladder and peritonitis. The other four patients succumbed after a brief illness; there was no history of memory disorder before their admission to the hospital, and none was present on initial examination, but the possibility of its having developed during the short terminal phase of the illness could not be excluded. The mammillary body lesions were chronic in nature in three of these five patients (including the one who was known for certain to be free of memory defect), and in all five cases the affection of the mammillary bodies was severe in degree.

A simliar case has been reported by Tsiminakis (1931). His patient was a serious alcoholic who was under observation for many years before his death from lobar pneumonia, and although he was of low intelligence, there was never any evidence of Korsakoff psychosis. Postmortem examination showed severe and probably long-standing degenerative changes confined to the mammillary bodies.

Clearly, on the basis of the foregoing observations, both our own and these of Tsiminakis, no connection can be made between mammillary body lesions and a Korsakoff amnesic defect. This conclusion is now supported by a considerable body of evidence, derived from clinical-pathologic studies of diseases other than the Wernicke-Korsakoff syndrome and from animal experimental studies. This evidence is reviewed in the following pages.

CLINICAL AND PATHOLOGIC OBSERVATIONS IN MAN

Evidence that lesions of the medial thalamus give rise to a persistent amnesic defect has been accumulating since the early 1950's. The early stereotactic procedures of Spiegel and Wycis and their colleagues (1955), in which lesions were placed in the medial-dorsal nuclei of 30 patients with severe emotional disturbances, were frequently associated with temporal disorientation, lack of spontaneity, and a memory defect for recent and past events which persisted for several weeks following operation.

Also, there are now on record numerous clinical-pathologic studies in which naturally occurring medial thalamic lesions of diverse causation have given rise to an enduring amnesic state. The most common lesions of this sort are medial thalamic infarctions, in the territory of the paramedian thalamoperforating arteries. These small vessels arise from the posterior cerebral arteries, between their point of origin (at the basilar bifurction) and the origins of the posterior communicating arteries. According to Percheron (1971), both medial thalamic regions may be supplied by branches of a single paramedian trunk, occlusion of which would account for bilateral infarction Guberman and Stuss (1983) described two patients with the abrupt onset of an amnesic state, among other neurologic signs, attributable to bilateral medial thalamic infarction (by CT scan) and have summarized the findings in 11 such cases in the literature. Neoplasms in the paramedian thalamic regions may have the same effects (see review of McEntee et al, 1976). Schulman (1957) described an unusual degenerative disease, confined to the thalamus and associated with a characteristic amnesic syndrome. A similar case had previously been described by Stern (1939). A detailed account of these and many other instances of impaired memory and learning due to medial thalamic lesions can be found in the review of Markowitsch (1982).

A shortcoming of all of these cases, from the point of view of clinical-pathologic correlation, is that in practically none were the lesions strictly confined to a particular thalamic nuclear mass and specifically to the medial-dorsal nucleus. This is true also of the cases of diencephalic amnesia in which bilateral or unilateral thalamic lesions have been demonstrated by CT scans. Even the noted patient of Squire and Moore (1979), who sustained a stab wound that appeared on CT scans to be limited to the left medial-dorsal nucleus, has, on recent MR imaging, been shown to have incurred more extensive (hypothalamic) damage (Squire LR, personal communication).

We have studied two cases of thalamic infarction that exemplify the difficulties of clinical-pathologic correlation in the non-alcoholic varieties of the diencephalic amnesic state. In both patients there had been the abrupt onset of an amnesic state which then persisted unchanged for several years, until the patients' death. In one patient, who eventually died of myocardial infarction old infarctions were found in each medial thalamus, the remainder of the brain being normal. On the right, the lesion was confined to the medial dorsal nucleus; this nucleus was involved on the left side as well but laterally the lesion encroached onto the ventrolateral nucleus. In the second patient, only the left (dominant) thalamus was involved. The major portion of the lesion was located in the medial dorsal nucleus, but inferolaterally the lesion involved the centromedian nucleus as well.

BEHAVIORAL ABNORMALITIES (?AMNESIC STATES) DUE TO THALAMIC LESIONS IN ANIMALS

Early experimental studies in animals also implicated the medial thalamus in learning and memory. Schreiner and associates (1953) observed that cats in which the medial dorsal nuclei were destroyed forgot their preoperative training and were very difficult to retrain. Similar observations in cats were made by Pechtel and his group (1955) and by Ingram (1958).

In the monkey, Chow (1954) found that bilateral lesions of the pulvinar, or of the pulvinar together with the medial dorsal nucleus, caused no apparent abnormality of learning or behavior; in none of these monkeys, however, did the lesions occupy more than 20 percent of the volume of the medial dorsal nucleus. Brierley and Beck (1958), on the basis of their observations in monkeys, concluded that for behavioral abnormalities to be long lasting, destruction of the medial dorsal nucleus must be bilateral and virtually complete. This view was substantiated by the careful experimental studies of Schulman (1964); he demonstrated a severe and enduring impairment of delayed response performance in three monkeys in which the medial dorsal nucleus had been completely destroyed bilaterally.

Among modern experimental studies that of Zola-Morgan and Squire (1985) is particularly noteworthy. They showed that small bilateral lesions, confined to the posterior portion of the medial dorsal nucleus, gave rise in monkeys to a markedly impaired performance on a behavioral test that has been used in recent years as a model of the human amnesic syndrome (*delayed non-matching to sample*). Moreover, the same monkeys showed no defect in learning pattern discrimination problems - a finding that is consistent with the preserved capacity of the Korsakoff patient to learn certain cognitive skills such as mirror-reading (Cohen and Squire, 1980).

EFFECTS OF EXPERIMENTAL ABLATION OF THE MAMMILLARY BODIES

There is no convincing experimental evidence that lesions of the mammillary bodies give rise to a defect in retentive memory. Always, in the older experimental work, there was the question whether certain learned responses or behavioral changes in the rat or other animals could be interpreted as a defect in retentive memory, comparable to that of Korsakoff psychosis in man. To compound the difficulty, lesions in the mammillary bodies and related structures may have varying effects, depending upon the particular learned response that is being tested. Thus, Thompson and Hawkins (1961) found that lesions of the mammillary bodies did not interfere with retention of avoidance-conditioned responses in rats; similar observations were made in cats (Dahl et al, 1962) and in monkeys (Ploog and MacLean, λ1963). Neither destruction of the mammillothalamic tract (Thompson and Massopust, 1960) nor of the mammillary bodies (Thompson and Hawkins, 1961) altered the postoperative retention of visual and kinesthetic discrimination habits. On the other hand, Thompson (1963) found that destruction of the mammillary bodies, mammillothalamic tracts, or anterior nuclei of the thalamus impaired the ability of the rat to make right-left discriminations in a T-maze, in order to avoid being shocked. Krieckhaus and Randall (1968) repeated Thompson's experiments, substituting thirst for escape from pain in training rats, and found no decrements in performance following bilateral lesions of the mammillothalamic tracts. They suggested that the decrements found by Thompson might be attributable to defects in motivation rather than cognitive function and to lesions that damaged parts of the thalamus continguous to the mammillothalamic tracts. The experiments of Ploog and McLean (1963), Dahl et al (1962), Woody and Ervin (1966) and Kim et al (1967) also negate the role of mammillary body lesions in producing disturbances in memory function; in nome of these studies was there evidence of a "recent memory" deficit following bilateral lesions of the mammillary nuclei.

More recent experimental data have helped to clarify the effect of mammillary body lesions on memory. The findings of Aggleton and Mishkin (1985) are particularly noteworthy in this respect insofar as they utilized monkeys that were trained on an object recognition task - delayed nonmatching to sample - a task that has proved to be sensitive to human amnesia. They found that monkeys with complete destruction of the medial mammillary nuclei showed little if any deficit in object recognition. At the same time, the damaged monkeys did show a significant impairment on a spatial discrimination task as well as on reversal of the discrimination. Appleton and Mishkin took these results to mean that mammillary body damage is in itself insufficient to produce a significant degree of amnesia, but that it does play some role in mnemonic integration. Holmes et al (1983) had also found that spatial learning (but not analogous, non-spatial reversal learning) is impaired in monkeys with mammillary body lesions. Other authors, notably Mahut and her colleagues (1971, 1972, 1973) had demonstrated a similar abnormality in monkeys subjected to bilateral lesions of the fornix or of the hippocampus. On the basis of these observations, Aggleton and Mishkin (1985) proposed that the mammillary body is an integral part of a hippocampal-fornix-mammillary body circuit that may contribute to, but is not essential for, recognition memory. They proposed further that the development of a full-blown amnesic syndrome requires the additional interruption of a parallel limbic circuit, specifically one involving the amygdala. Interestingly these authors invoked our neuropathologic findings (see above) to bolster their proposal, pointing out that in five of our Wernicke patients *without* amnesia, the lesions were confined to the mammillary bodies, whereas in 38 such patients *with* amnesia there was damage to both the mammillary bodies and the medial-dorsal nucleus of the thalamus. The notion that a combined lesion (mammillary bodies and medial thalami) is necessary to produce Korsakoff psychosis may have validity in non-human primates. In *man*, however, lesions confined to the medial thalami are quite sufficient to produce a profound and persistent amnesic state, as was fully discussed above.

ROLE OF FORNIX LESIONS IN THE CAUSATION OF AN AMNESIC STATE

The mammillary bodies have also been implicated in memory function by virtue of their anatomic connection with the hippocampal formations, via the fornix. However, to think of the anatomy of memory merely in terms of an interruption of this pathway, as Barbizet (1963) has done, is an over-simplification, to say the least. Bilateral interruption of the anterior pillars of the fornix has not been followed by a permanent memory defect in man (Dott, 1938; Garcia-Bengochea et al, 1954; Squire and Zola-Morgan, 1983), the patient reported by Sweet et al (1959) not withstanding. The latter authors described a patient who was left with a permanent amnesic state following section of the fornices and removal of a third ventricular tumor. It should be pointed out that the memory quotient in this patient (which was only tested postoperatively) was in the low normal range, and fell only 13 points below the intelligence quotient (usually, in the Korsakoff amnesic state, the difference between the IQ and MQ is much greater). The disturbance of memory in this patient may have resulted from damage to diencephalic structures, either from compressive effects of the tumor or its removal, rather than from section of the fornices. Also, in monkeys bilateral lesions of the fornix have not resulted in a defect of learning or memory, as closely as this can be judged by behavioral tests (Garcia-Bengochea et al, 1951: Squire and Zola-Morgan, 1983).

Although the fornix constitutes the main efferent bundle from the hippocampus to the mammillary body, it needs to be remembered that some of the fornical fibers originate in structures other than the hippocampus; a modest contribution to the fornix is made by the cingulate gyrus and the indusium griseum, and a more important one, by the septal area and hypothalamus (Nauta, 1956, 1958; Valenstein and Nauta, 1959). Furthermore, evidence derived from animal experiments indicates that fornical fibers project to many parts other than the mammillary bodies. Daitz and Powell (1954) found that most of the large cells in the medial septal nucleus and some of the large cells in the nucleus of the diagnonal band of Broca degenerate following sectioning of the fimbria of the fornix. Nauta (1956, 1958) has shown, in both the rat and the cat, that fibers from the hippocampal formation project to the septal area, preoptic hypothalamuc, dorsal and periventricular regions of the hypothalamus, anterior nucleus of the thalamus, median and paramedian regions of the thalamus, supramammillary region and rostral portion of the central gray matter of the midbrain. An even more extensive projection of the hippocampal formation- fornix system to basal forebrain, preoptic, and hypothalamic areas has been demonstrated in the squirrel monkey (Poletti and Creswell, 1977).

SUMMARY

This discussion has been concerned with one small corner of the anatomy of memory, namely, with the role of mammillary body lesions in the causation of the Korsakoff amnesic state. Clinical-pathologic studies, both our own (of alcoholics with the Wernicke-Korsakoff syndrome) and those of others (in patients with thalamic infarctions and neoplasms) provide unambiguous evidence that medial thalamic lesions. without additional lesions of the mammillary bodies, are quite sufficient to produce a severe and enduring amnesic state. Behavioral abnormalities in animals, insofar as they can be equated with a defect in retentive memory, tend to substantiate the observations in humans.

RESUMEN

Este estudio se refiere a un pequeño sector de la anatomía de la memoria en especial el papel de las lesiones del cuerpo mamilar como causa del estado amnésico Kor

sakoff. Estudios clínico-patológicos, ambos realizados por nosotros (de alcohólicos con el síndrome Wernicke-Korsakoff) y aquellos otros (en pacientes con infartos y neoplasmas talámicos) brindan una clara evidencia que lesiones talámicas internas sin lesiones adicionales de los cuerpos mamilares, son suficientes para originar un estado amnésico severo y persistente. Anormalidades de conducta en animales tales que pueden ser vinculadas con un defecto en la retención de memoria, substancian las observaciones en seres humanos.

RÉSUMÉ

Cette discussion ne concerne qu'une partie limitée de l'anatomie de la mémoire, celle du rôle des lésions du corps mamillaire à l'origine du syndrome de Korsakoff. Les études anatomo-pathologiques, aussi bien les nôtres (de syndromes de Wernicke-Korsakoff chez des alcooliques) que celles d'autres auteurs (lésions thalamiques vasculaires ou néoplasiques) apportent sans conteste la preuve que des lésions thalamiques médianes, sans association de lésions des corps mamillaires, sont suffisantes pour réaliser un état amnésique durable. Les anomalies comportementales chez l'animal, pour autant qu'elles puissent être l'équivalent d'un déficit de la rétention mnésique conduisent à confirmer les observations faites chez l'homme.

ZUSAMMENFASSUNG

Diese Untersuchung bezieht sich auf einen kleinen Abschnitt der Anatomie des Gedächtnisses insbesonders auf die Rolle von Läsionen des corpus mamilare als Ursache des Zustandes Korsakoff'scher Amnesie. Klinisch-pathologische Untersuchungen (beide von uns durchgeführt) von Alkoholikern mit Wernicke-Korsakoff'schem Syndrom und jenen anderen bei Patienten mit Infarkten und Geschwülsten im Thalamus liefern einen klaren Augenschein dafür, dass Läsionen innerhalb des Thalamus ohne zusätzliche Läsionen der corpora mamilaria genügen, um einen schweren und dauernden amnestischen Zustand zu verursachen. Solche Verhaltensabnormalitäten bei Tieren, die in Verbidung sein können mit einer Störung im Bewahren des Gedächtnisses, verdeutlichen die Beobachtungen beim Menschen.

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Hormonal and Neurotransmitter Interactions in the Modulation of Memory Storage: Involvement of the Amygdala

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INTRODUCTION

There is extensive evidence indicating that memory storage is modulated by a variety of hormones that are normally released by midly arousing stimulation. (de Wied, 1984; McGaugh and Gold, in press; McGaugh, 1983). Our current work focuses on the central mechanisms underlying the effects, on memory modulation, of treatments affecting adrenergic and opiate receptor systems. Our findings strongly suggest that treatments affecting adrenergic receptor systems as well as opiate receptor systems modulate memory through influences involving noradrenergic receptors in the amygdala. We will first review some of our findings concerning the involvement of adrenergic and opioid peptide systems in the modulation of memory storage and then discuss the interaction of adrenergic and opioid peptide systems in their influences on noradrenergic receptors within the amygdala.

EPINEPHRINE EFFECTS ON MEMORY

Numerous studies have reported that, in rats and mice, systemic administration of the adrenal medullary hormone epinephrine immediately following a training session alters subsequent retention of the response (Gold and van Buskirk, 1976; Borrell, de Kloet, Versteeg, and Bohus, 1983; McGaugh and Gold, in press). Since epinephrine is known to be releassed from the adrenal medulla by the kind of aversive stimulation typically used in animal memory experiments (Gold and McCarty, 1981) the findings have been interpreted as supporting the view that endogenously-released epinephrine has effects which serve to modulate the storage of recently acquired information (Gold and McGaugh, 1975).

If it is assumed that epinephrine effects on memory are due to a general modulating effect on memory storage, then the effects should not be restricted to any particular learning task. There is clear evidence that posttraining administration of epinephrine affects retention of both active and inhibitory avoidance tasks (Gold and van Buskirk, 1976; Liang, Bennett; and McGaugh, 1985) as well as discrimination tasks where choice rather than response latency is used to assess retention. For example, in a recent study we found that posttraining epinephrine alters subsequent long-term retention of a footshock-motivated visual discrimination task (Introini-Collison and Mc Gaugh, 1986). Mice in this study were trained to escape from a mild footshock by entering the left, lighted arm of a Y-maze (criterion of 3 successive correct choices) and were then given posttraining injections of either saline or epinephrine (0.3 or 1.0 mg/kg ip). On retention tested 1 day, 1 week, or 28 days later, the location of the correct alley was reversed and the mice were given 6 trials on which they could escape from the shock only by entering the right, dark alley. As Figure 1 shows, at each retention interval the mice given a low dose of epinephrine made more errors than did saline controls on the reversal training, while mice given the high dose made fewer errors. It is particularly important to

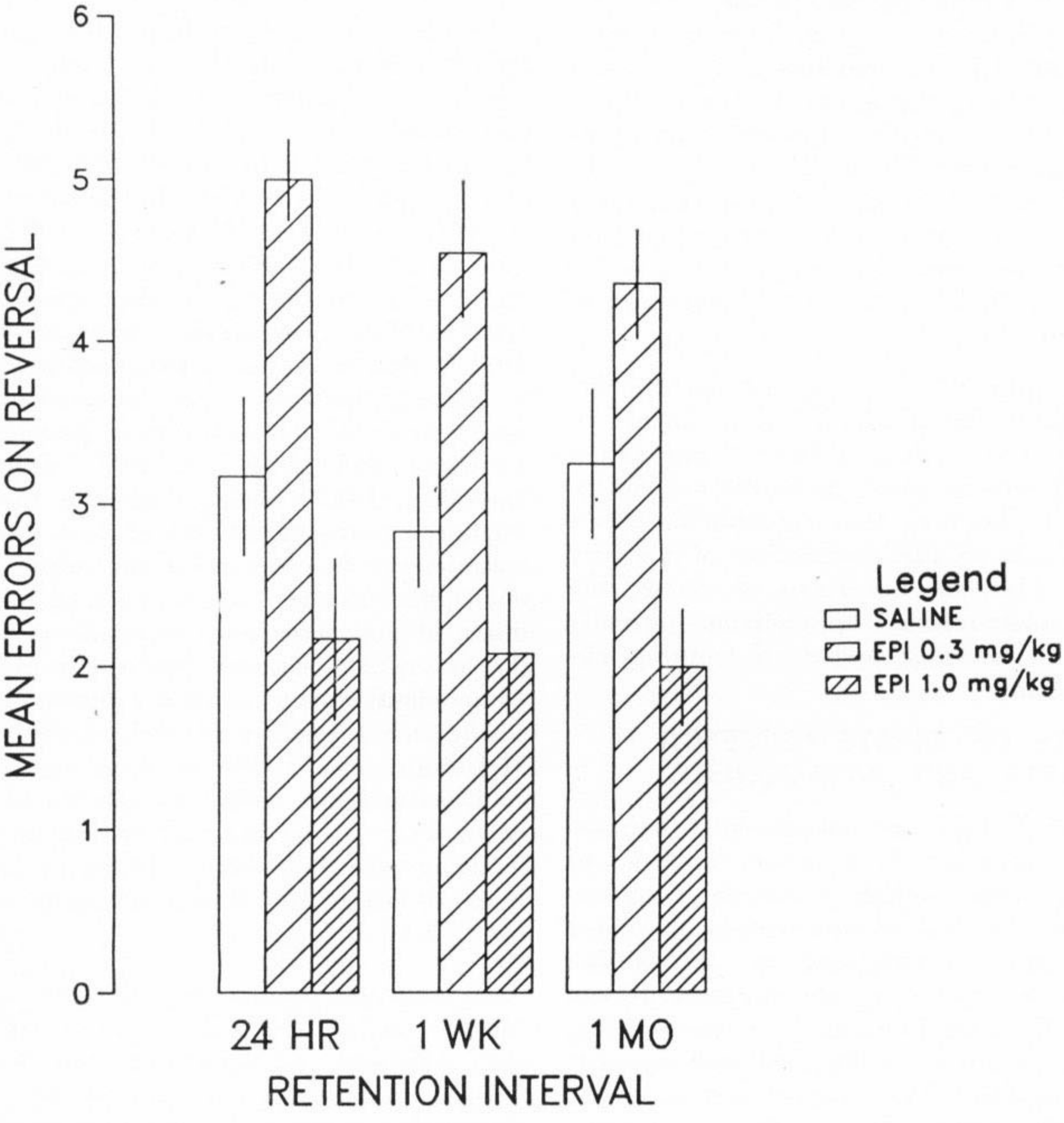

Fig. 1. — *Effects, in mice, of immediate posttraining epinephrine (ip) on discrimination reversal training 1 day, one week, or one month following original training (Mean ± S.E.). Retention of the original discrimination is indexed by errors made on the discrimination reversal. N=18 for 1 day group and N=12 for one week and one month groups. (From Introini-Collison and McGaugh, 1986).*

note that the memory-modulating effects of a single posttraining injection of epinephrine are long-lasting: Effects assessed on the one-month retention test were comparable to those seen one-day following training.

We interpret these findings as indicating that the low dose of epinephrine enhanced retention of original discrimination while the high dose of epinephrine impaired retention of the original discrimination. These findings are highly comparable to those that we and other investigators have obtained with inhibitory avoidance tasks (Gold and van Buskirk, 1976; Introini-Collison and McGaugh, 1986). Further, while the effects of epinephrine on memory have been typically studied in experiments using aversive motivation, we have found that epinephrine can also influence the retention of an appetitively-motivated discrimination task. In both appetitively and aversively motivated training tasks the effects of epinephrine on retention are blocked by systemically administered α- and β-antagonists. (Sternberg *et al.*, 1985; 1986).

Our findings indicating that epinephrine has highly simliar effects in a variety of learning tasks using different motivation and different response measures are consistent with the view that epinephrine has a general role in the modulation of memory storage. This does not mean, of course, that endogenously-released epinephrine normally modulates learning under all training circumstances.

EFFECTS OF OPIATE RECEPTOR AGONISTS AND ANTAGONISTS

There is also extensive evidence that memory, as assessed in a variety of types of training tasks, including aversively-motivated tasks as well as appetitively-motivated tasks, can be modulated by posttraining treatments affecting opiate receptors. Retention is generally impaired by posttraining administration of opiate receptor agonists (e.g. morphine, β-endorphin) and enhanced by opiate receptor antagonists such as naloxone and naltrexone (Castellano, 1975; 1981; Gallagher, 1985; Introini-Collison and Baratti, 1986; Izquierdo, 1979; Messing, *et al*, 1979). The memory-enhancing effects of opiate receptor antagonists appear to be based on central effects, since retention is not affected by posttraining i.p. administration of naltrexone methyl bromide (MR2263), an opiate receptor antagonist that does not readily pass the blood-brain barrier. Further, the memory impairing effects of β-endorphin as well as morphine also appear to be centrally mediated since MR2263 does not antagonize the effects of these treatments (Introini, McGaugh, and Baratti, 1985).

INTERACTION OF ADRENERGIC, NORADRENERGIC AND OPIATE SYSTEMS

Evidence from several studies suggests that epinephrine effects on retention may involve interactions with a brain opioid peptide system. For example the findings that the memory-impairing effect of high doses of epinephrine is blocked by naloxone (Introini-Collison and McGaugh, 1987; Izquierdo and Dias, 1983) are consistent with other evidence indicating that epinephrine releases brain β-endorphin (Carrasco et al., 1982). However, the finding that low doses of epinephrine block the memory-impairment produced by posttraining β-endorphin (Introini and McGaugh, 1987; Izquierdo and Dias, 1985) clearly indicates that the memory-enhancing effects of low doses of epinephrine do not involve the release of β-endorphin. Further, we have found that low doses of naloxone and epinephrine which do not affect memory when administered alone significantly enhance memory when administered together (Introini-Collison and McGaugh, 1987). As is discussed below, such an additive effect is expected in view of evidence suggesting that the memory-modulating effect of both of these treatments involves the release of central norepinephrine (NE).

Opiates and opioid peptides are known to exert an inhibitory influence on NE neurons (Walker, Khachaturian and Watson, 1984). Electrophysiological studies have shown that systemically as well as iontophoretically administered opiate receptor agonists induce naloxone-reversible depression of spontaneous as well as stimulation-induced firing rates of locus coeruleus neurons

(Bird and Kuhar, 1977; Korf, Bunney and Aghajanian, 1974; Pepper and Henderson, 1980; Young, Bird and Kuhar, 1977; Strahlendorf, Strahlendorf, and Barnes, 1980). Morphine and β-endorphin depress the release of NE from rat cerebral cortex slices (Arbilla and Langer, 1978; Montel, Starke, and Weber, 1974). Further, infusion of morphine into the cortical terminal fields of locus coeruleus neurons depresses excitability to direct stimulation (Nakamura *et al.*, 1982). The finding that opiate receptor binding is reduced in NE terminal regions following 6-OHDA destruction of NE cell bodies suggests that opiate receptors may be located presynaptically on NE cells (Llorens *et al.*, 1978). Tanaka and his colleagues (Tanaka, *et al.*, 1982) have reported that stress increases NE turnover in the amygdala and that the turnover is enhanced by naloxone. These findings suggest that endogenous opioid peptides in the amygdala attenuate stress-induced NE turnover.

The findings of several studies suggest that adrenergic as well as opioid peptidergic systems may interact with central NE in their effects on memroy. Izquierdo and Graudenz' (1980) finding that propranolol (a $\beta_{1,2}$ antagonist) blocked the enhancing effects of naloxone on memory suggest that opioid antagonists may enhance retention by releasing brain NE neurons from the inhibitory effects of opioid peptides. Other recent evidence has provided additional support for the view that opioid effects on memory involve NE. For example, Introini-Collison and Baratti (1986) reported that naloxone potentiates the memory-enhancing effects of the centrally acting β-adrenergic agonist clenbuterol. Gallagher and her colleagues have reported that 6-OHDA lesions of the dorsal noradrenergic bundle block the enhancing effects of naloxone on retention (Fanelli, Rosenberg, and Gallagher, 1985; Gallagher, Rapp and Fanelli, 1985). Further, Introini-Collison and Baratti (1986) have shown that the enhancing effects of naloxone on memory are also blocked in animals treated with DSP4, a relatively specific noradrenergic neurotoxin. The findings that the memory-enhancing effects of naloxone are not blocked by a peripherally-acting β-antagonist (sotalol), or by α-antagonists (phenoxybenzamine or phentolamine) argue that naloxone effects on memory may specifically involve central β-adrenergic receptors (Introini-Collison and Baratti, 1986).

INVOLVEMENT OF THE AMYGDALA

While the findings of studies using noradrenergic neurotoxins and systemically-administered β-receptor antagonists suggest that naloxone effects on memory require an intact NE system, they do not reveal whether NE in any specific brain region is of particular importance. Other recent findings suggest that effects of naloxone and epinephrine on memory may involve the activation of NE receptors within the amygdala. Extensive anatomical evidence suggests that the amygdaloid complex is a likely candidate for a locus of the effects of treatments affecting opiate and noradrenergic receptors. Noradrenergic receptors are found in several amygdala nuclei (U'Prichard *et al.*, 1980), particularly in the central and basolateral nuclei (Fallon, 1981). Opiate receptors are also found throughout the amygdaloid complex. Enkephalin fibers and cell bodies are located in the central nucleus and β-endorphin is located in the medial and central nuclei (Bloom and McGinty, 1981).

Many studies have shown that posttraining low intensity (sub-seizure) electrical stimulation can modulate retention (Kesner, 1982; McGaugh and Gold, 1976). In view of evidence that the effects of amygdala stimulation on retention are blocked by lesions of the stria terminalis (ST), a major amygdala pathway, (Liang and McGaugh, 1983b) it seems likely that amygdala stimulation affects retention by modulating storage processes at sites in other brain regions. Our view that epinephrine affects memory through influences involving the amygdala is supported by the finding that lesions of the ST also block the effects of epinephrine on memory (Liang and McGaugh, 1983a). More recently, as is shown in Figure 2, we have found that lesions of the ST also block the memory-

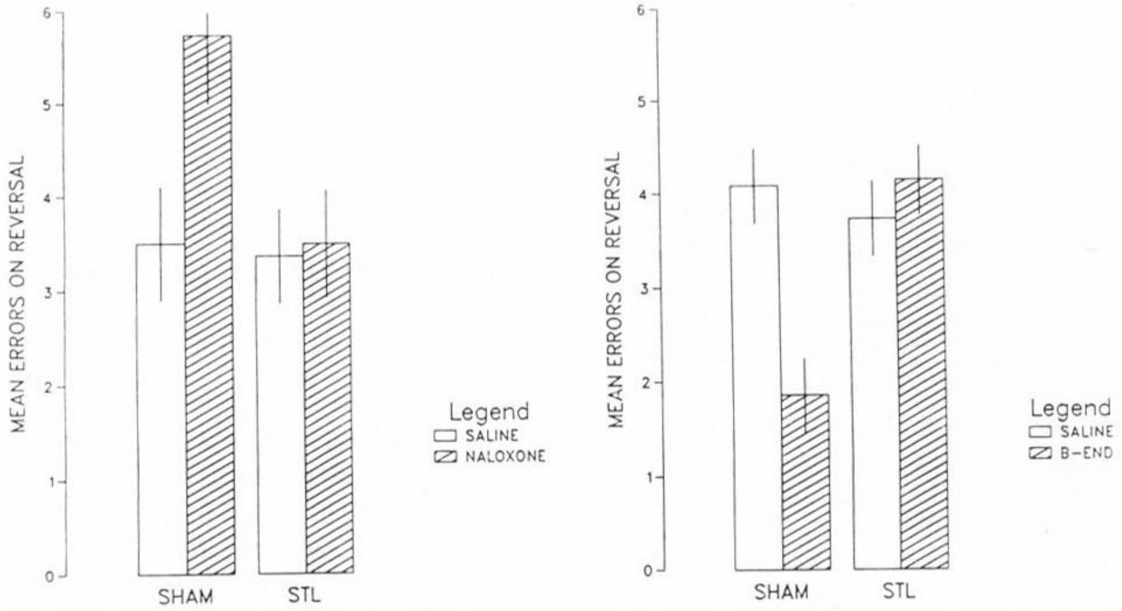

Fig. 2. — *Effects of systemic naloxone (left) or β-endorphin (right) on one-week retention of the visual discrimination task in rats with sria terminalis lesion (From McGaugh, Introini-Collison, Juler and McGaugh, 1986).*

enhancing effects of posttraining peripheral injections of naloxone as well as the memory-impairing effects of β-endorphin (McGaugh, Introini-Collison, Juler, and Izquierdo, 1986). It seems unlikely that the amygdala is a site of memory storage in view of previous evidence from our laboratory (Liang *et al.*, 1982) indicating that bilateral lesions of the amygdala do not impair retention if they are produced several days following training. However, such lesions do impair retention if produced within a few days after training. This evidence argues that the amygdala is involved in modulating the storage of recently acquired information.

INVOLVEMENT OF INTRA-AMYGDALA NOREPINEPHRINE

While it is generally assumed that epinephrine does not pass the blood-brain barrier, the route by which epinephrine affects the brain is not yet known. One possibility is that the effects of peripheral epinephrine may be mediated by activation of visceral afferents projecting to central noradrenergic systems which are known to the amygdala (via the ST and the ventral amygdalo-fugal pathway).

If modulation of memory storage processes involves noradrenergic activation within the amygdala, then it should be possible to influence retention with posttraining intra-amygdala injections of noradrenergic agonists and antagonists. There is extensive evidence to support this implication. For example, Gallagher and her colleagues have reported that retention of an inhibitory avoidance response is impaired by posttraining intra-amygdala injections of the β-antagonists, propranolol and alprenolol. The effect is time-dependent, stereo-specific and is attenuated by concurrent intra-amygdala administration of NE (Gallagher, *et al.*, 1981). In addition, Kesner and his colleagues found that posttraining intra-amygdala injections of NE (in high doses) can impair retention (Ellis and Kesner, 1981, 1983; Ellis, Berman, and Kesner, 1983).

Studies from our laboratory have shown that retention is enhanced by low doses of NE administered intra-amygdally posttraining (Liang, Juler and McGaugh, 1986). The effect of NE was blocked by concurrent intra-amygdala injections of propranolol. Further, as is shown in Figure 3, posttraining intra-amygdala injections of propranolol blocked the retention-enhancing effects of *peripherally* administered epinephrine. This finding provides strong evidence for

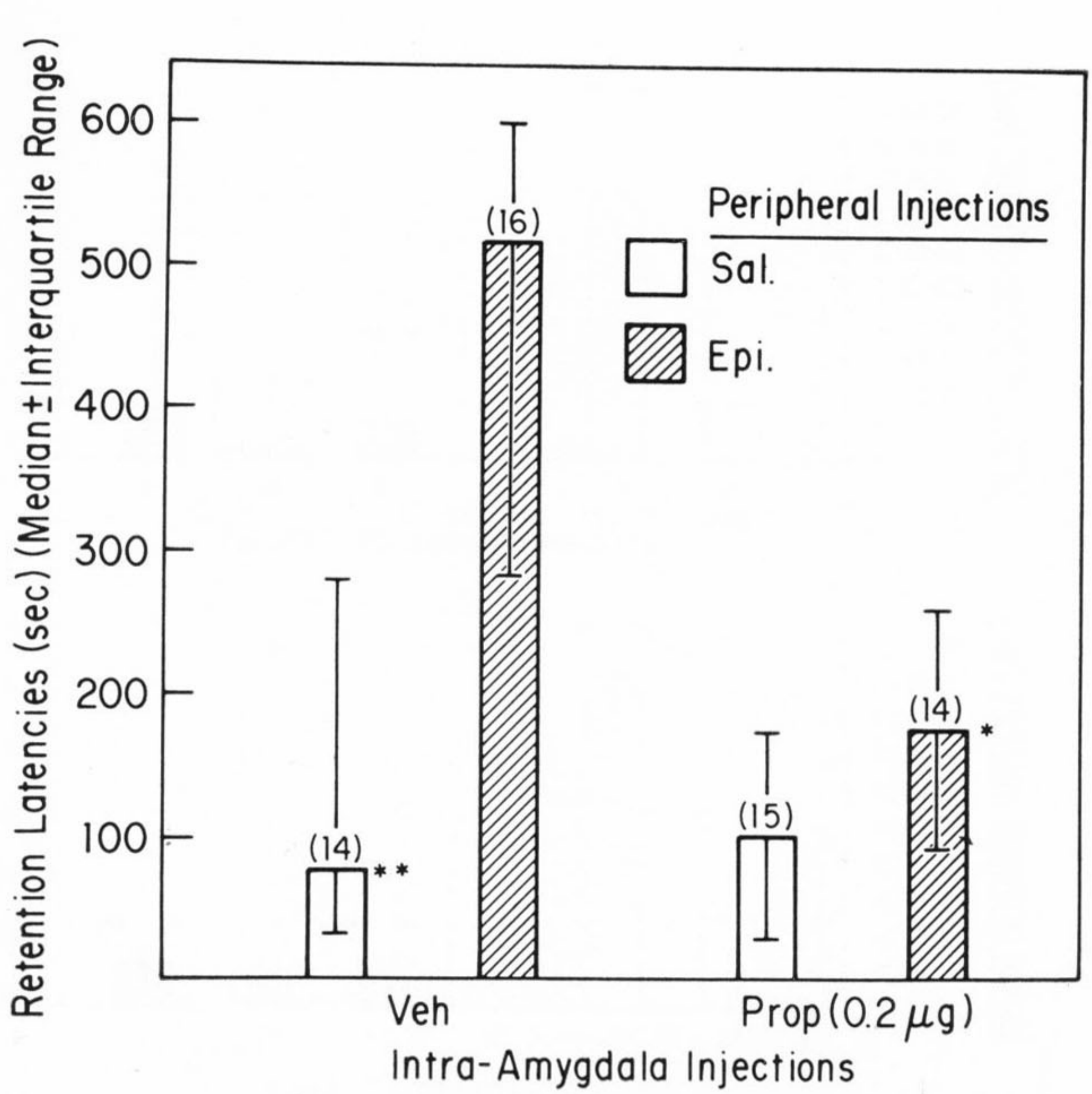

Fig. 3. — *Effects of posttraining intra-amygdala administration of propranolol and ip administration of epinephrine on retention of an inhibitory avoidance response (Median [interquartile range]). N's shown in parentheses. * P<0.05 ** P<0.02 compared with group given vehicle intraamygdally and epinephrine ip. (From Liang, Juler and McGaugh, 1986).*

our view that epinephrine affects memory by influencing the release of NE within the amygdala.

INTERACTION OF NORADRENERGIC AND OPIATE SYSTEMS

Retention can also be modulated by intra-amygdala injections of opioid agonists and antagonists. Gallagher and her colleagues reported that posttraining intra-amygdala injections of the opiate agonist levorphanol produced a naloxone-reversible retention deficit and that posttraining intra-amygdala naloxone administered alone enhanced retention (Gallagher and Kapp, 1978; Gallagher *et al.*, 1981). In further support of the interpretation that effects of opiate receptor antagonists on memory may be due to blocking of opioid inhibition of NE release within the amygdala, Gallagher, Rapp and Fanelli (1985) reported that 6-OHDA lesions of the dorsal noradrenergic bundle block the effects of posttraining intra-amygdala injections of naloxone on retention.

Other recent findings from our laboratory provide additional evidence suggesting that the modulating effects of naloxone on re-

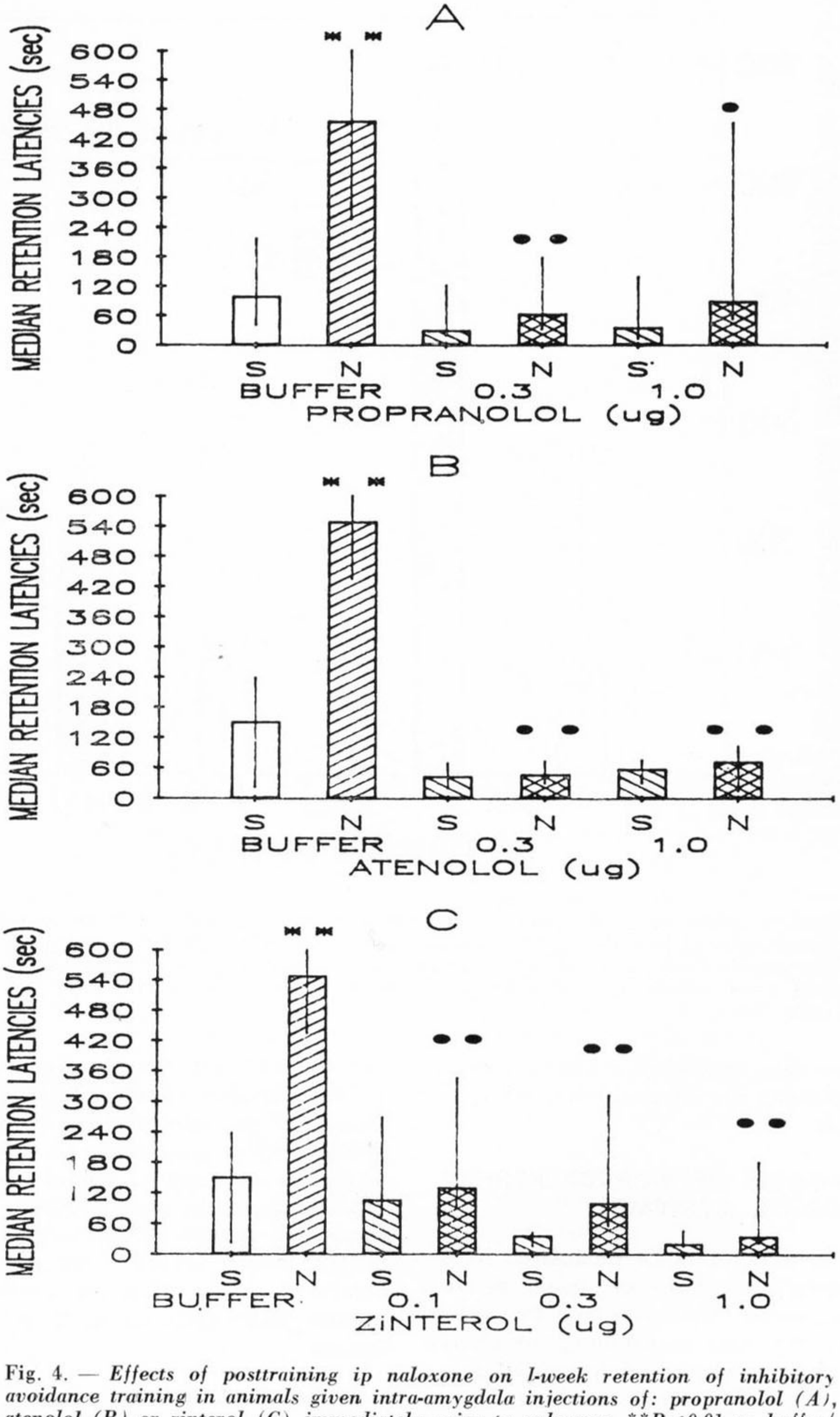

Fig. 4. — *Effects of posttraining ip naloxone on l-week retention of inhibitory avoidance training in animals given intra-amygdala injections of: propranolol (A), atenolol (B) or zinterol (C) immediately prior to naloxone.* $^{**}P<0.01$ *vs buffer/saline control group.* °$P<0.05$ *and* °°$P<0.01$ *vs buffer/naloxone-injected group (From McGaugh, Introini-Collison and Nagahara, in press).*

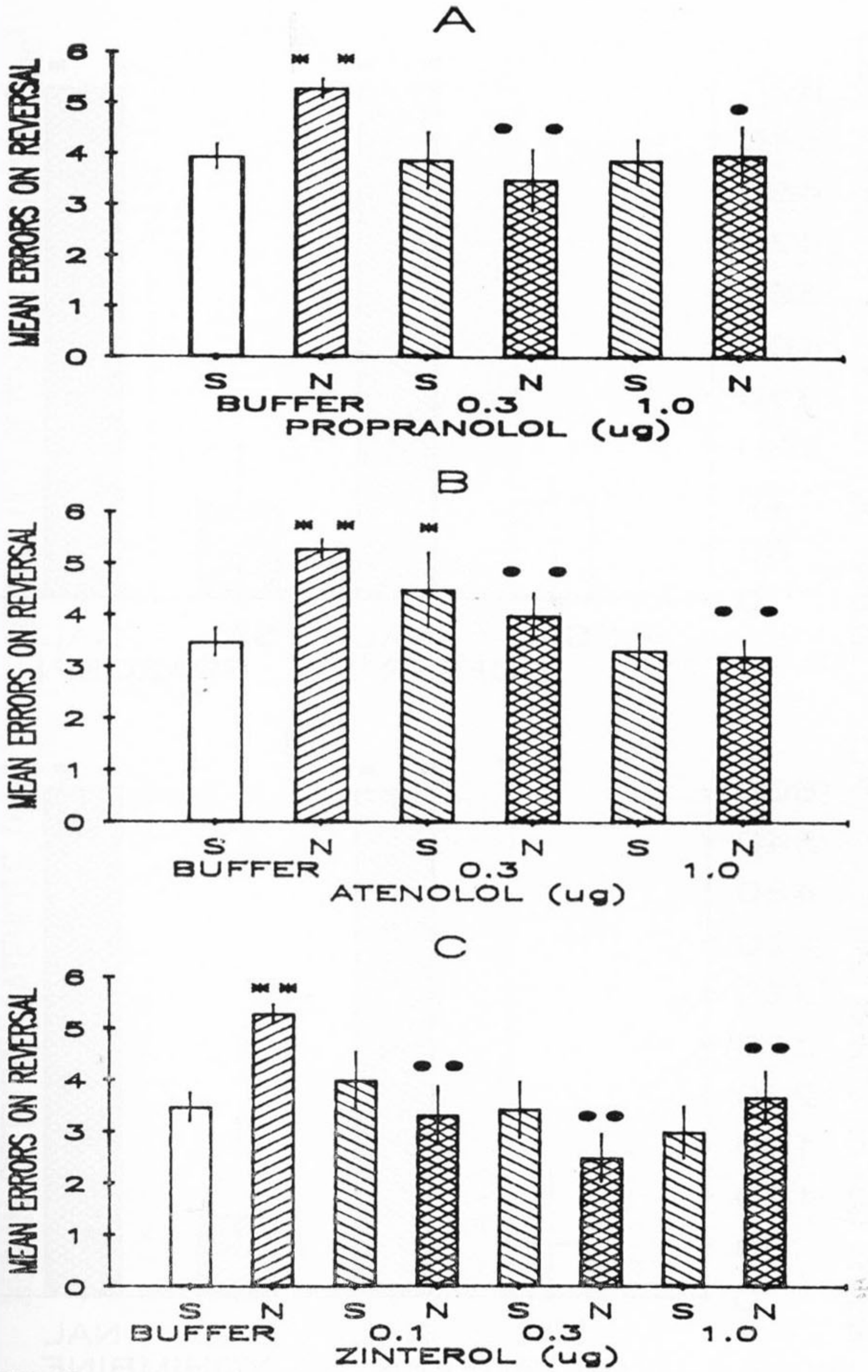

Fig. 5. — *Effects of posttraining ip naloxone on 1-week retention of Y-maze discrimination training in animals given intra-amygdala injections of propranolol (A), atenolol (B) or zinterol (C) immediately prior to naloxone.* $*P<0.05$ *and* $**P<0.01$ *vs buffer/saline-injected control group.* $°P<0.05$ *and* $°°P<0.01$ *vs buffer/naloxone injected control group (From McGaugh, Introini-Collison and Nagahara, in press).*

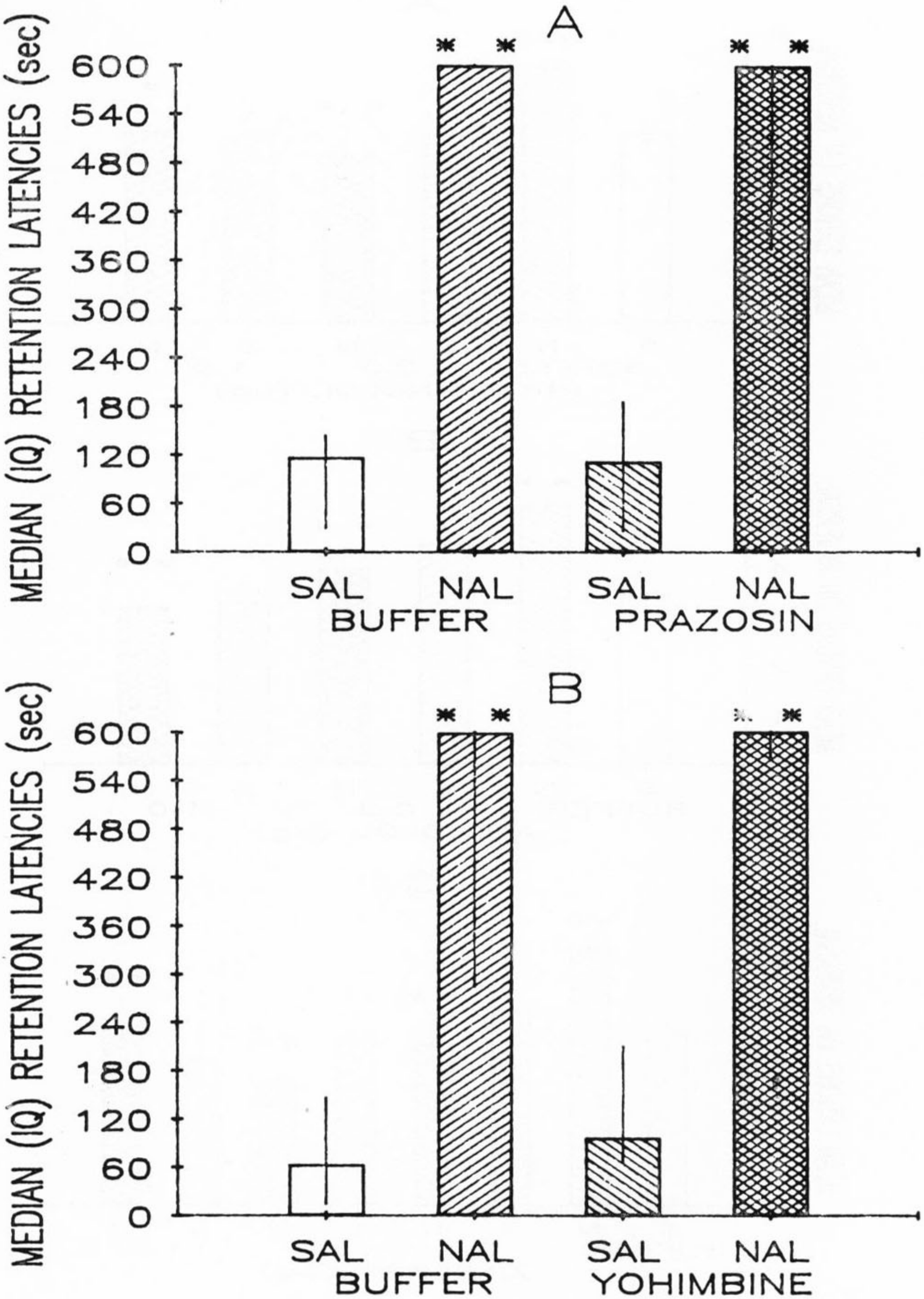

Fig. 6. — *Effects of posttraining ip naloxone on 1-week retention of inhibitory avoidance task in animals given intra-amygdala injections of prazosin (A) or yohimbine (B) immediately prior to naloxone injection. ** $P<0.01$ vs saline-injected control group (From McGaugh, Introini-Collison and Nagahara, in press).*

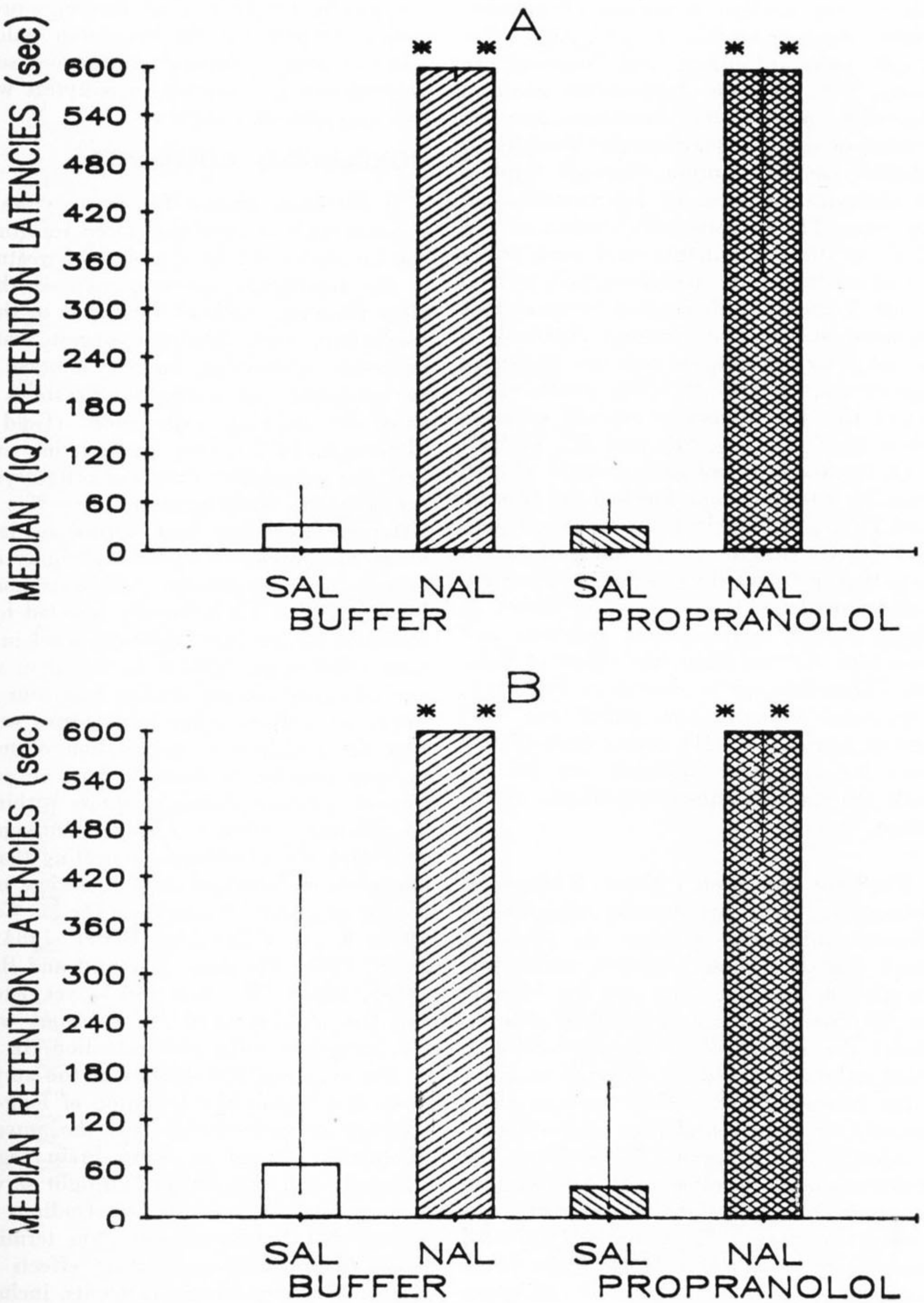

Fig. 7. — *Effects of posttraining ip naloxone on 1-week retention of the inhibitory avoidance task in animals given intra-caudate (A) or intracortex (B) propranolol immediately prior to naloxone injection. ** $P<0.01$ vs saline-injected control group (From McGaugh, Introini-Collison and Nagahara, in press).*

tention may involve activation of noradrenergic receptors within the amygdala (McGaugh, Introini-Collison, and Nagahara, in press). Rats in these experiments received immediate posttraining intra-amygdala injections of α- and β-adrenergic blockers or a buffer control solution, through implanted cannulae, followed by i.p. injections of naloxone. The experiments examined the effects of these treatments on 1-week retention of an inhibitory avoidance task as well as our Y-maze discrimination reversal task discussed above. The findings obtained in the inhibitory avoidance task are shown in Figures 4A, 4B and 4C. The results obtained in the discrimination reversal task are shown in Figure 5A, 5B, and 5C. In both tasks, the memory enhancing effect of posttraining naloxone was blocked by propranolol (a $\beta_{1,2}$ antagonist), atenolol (a β_1 antagonist) or zinterol (a β_2 antagonist), in doses that did not affect retention when administered alone. However, as is shown in Figure 6, the α-antagonists prazosin and yohimbine did not block the effect of naloxone. Moreover, as is shown in Figure 7, propranolol injected into either the (A) caudate nucleus or (B) cortex immediately above the amygdala injection site did not block the memory-enhancing effects of naloxone.

We have interpreted these findings as indicating that peripherally-administered naloxone influences memory by blocking opioid peptide receptors located within the amygdaloid complex. Thus, on the assumption, as discussed above, that opioid peptides inhibit the release of NE, systemically-injected naloxone would be expected to induce the release of NE within the amygdala. Recently we have found that intra-amygdala injections of propranolol also block the memory-enhancing effects of posttraining intra-amygdala injections of naloxone. As is shown in Figure 8, propranolol injected immediately posttraining blocked the effect of naloxone on retention of the inhibitory avoidance task (8A) as well as a the Y-maze discrimination task (8B). As in the experiments using peripherally-injected naloxone, retention was tested 1-week following training (unpublished findings). Thus, considered together, our findings provide strong support for the view that naloxone affects memory through influences selectively involving β-adrenergic receptors within the amygdaloid complex.

CONCLUDING COMMENTS

It has been known for many years that the retention of newly-acquired information can be modulated by a variety of treatments if the treatments are administered shortly after training. And, as we noted in the introduction, such findings suggest that the processes underlying memory storage may be modulated by endogenous systems activated by training experiences (Gold and McGaugh, 1975). Our research has examined the possibility that hormonal systems activated by training may serve this role. Extensive evidence from numerous laboratories has indicated that retention is influenced by posttraining administration of hormones that are normally released by stimulation of the kind typically used in training (McGaugh, 1983). In addition, as reviewed above, recent studies from our laboratory as well as other laboratories suggest that the endogenous modulation of memory may involve, at least in part, an activation of norepinephrine receptors within the amygdaloid complex. These findings fit well with other evidence indicating that the amygdala is involved in the storage of recently acquired information (e.g., Cohen, 1975; Kapp, Pascoe and Bixler, 1984; LeDoux, 1986; Mishkin, Malamut and Bachevalier, 1984). We have not as yet determined how activation of NE receptors within the amygdala influences retention. In view of the evidence that lesions of the amygdala do not impair the retention of long-term memory it seems likely that the amygdala modulates storage in other brain regions. Alternatively, the amygdala might serve as a temporary storage site. Our findings indicating that lesions of the stria terminalis block the memory-modulating effects of a variety of posttraining treatments, including brain stimulation, epinephrine, naloxone and β-endorphin seem most consistent with the view that the amygdala is part of a system which serves to modulate memory storage at sites in other brain regions.

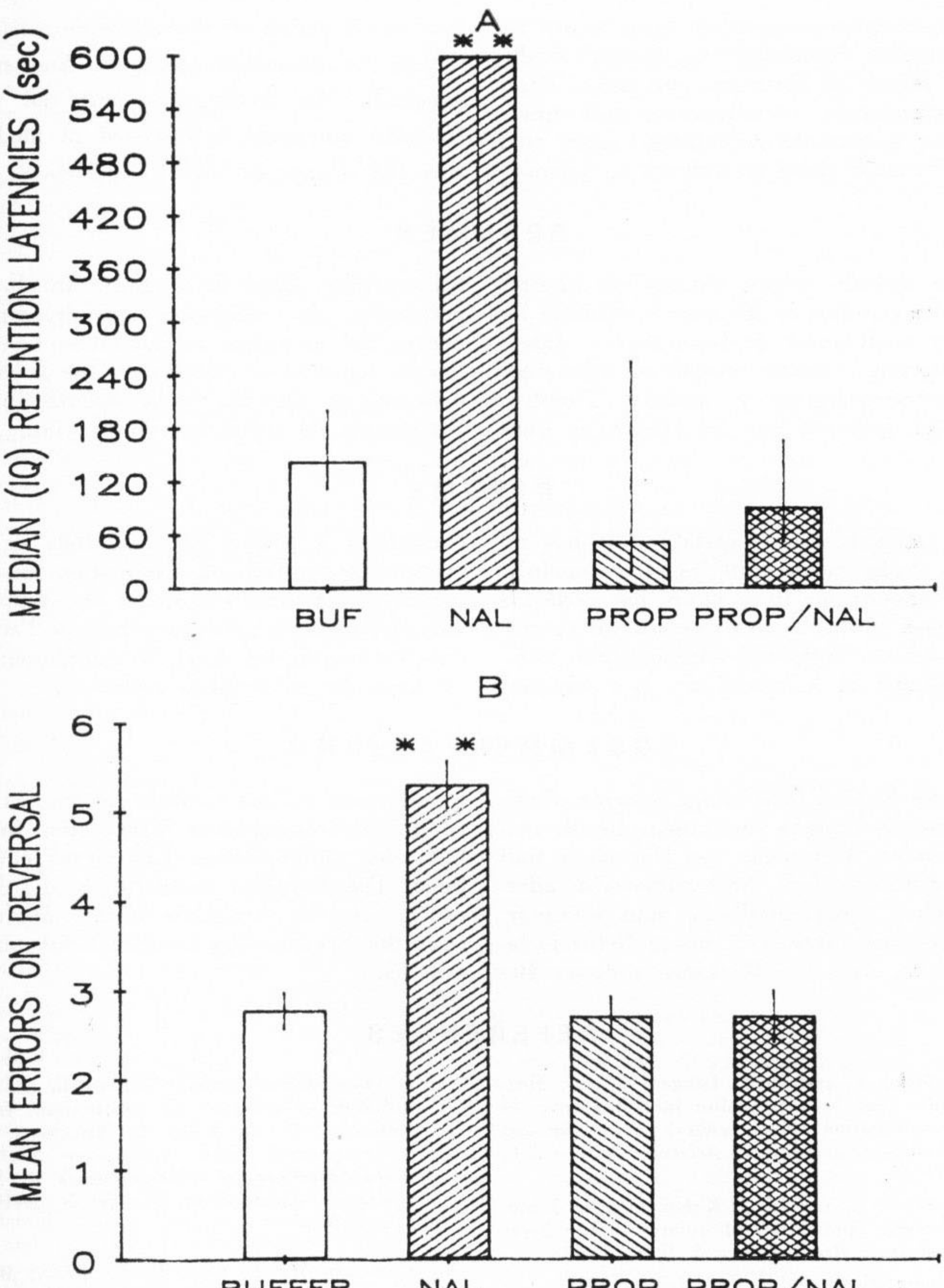

Fig. 8. — *Effects of posttraining intra-amygdala naloxone on 1-week retention of inhibitory avoidance (A) and Y-maze discrimination (B) training in animals given intra-amygdala injectons of propranolol immediately prior to naloxone. ** $P<0.01$ vs buffer-injected control group (unpublished findings).*

SUMMARY

This chapter summarizes some of our recent studies examining the memory-modulating effects of hormones and drugs affecting adrenergic, noradrenergic and opiate receptor systems. Our findings suggest that the effects of these treatments on memory involve activation of noradrenergic receptors within the amygdaloid complex. And, more generally, the findings support the view that the amygdala is involved in modulating the storage of recent experiences.

RESUMEN

Este capítulo resume algunos de nuestros recientes estudios en los que se examina los efectos modulantes de hormonas y drogas que afectan sistemas receptores, adrenérgicos, noradrenérgicos y opiáceos. Nuestros hallazgos sugieren que los efectos de estos tratamientos sobre la memoria involucran activación de receptores noradrenérgicos dentro del complejo amigdaloide. Los hallazgos soportan el criterio de que la amígdala está involucrada en la modulación del almacenaje de recientes experiencias.

RÉSUMÉ

Ce chapitre résume certaines de nos récentes études concernant les effets modulatoires obtenus sur la mémoire par certaines hormones et différentes drogues interessant les systèmes récepteurs adrénergique, noradrénergique et morphinique. Nos résultats conduisent à penser que les effets de ces traitements entrainent l'activation des récepteurs noradrénergiques du complexe amygdalien. Plus généralement, que l'amygdale est impliquée dans la modulation du stockage des informatios récentes.

ZUSAMMENFASSUNG

Dieses Kapitel fasst einige unserer jüngsten Untersuchungen zusammen, die die modulierenden Wirkungen von Hormonen und Drogen untersuchen, die rezeptorische, adrenergische, noradrenergische und opiumartige Systeme angreifen. Unsere Befunde legen nahe, dass die Wirkungen dieser Behandlungen auf das Gedächtnis die Aktivierung noradrenergischer Rezeptoren innerhalb des amigdaloiden Komplexes einschliesst. Die Befunde unterstützen die Meinung, dass die Amigdala in der Modulierung der Speicherung frischer Erfahrungen mitwirkt.

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Age-Associated Memory Impairment: Diagnostic Criteria and Treatment Strategies

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The older, healthy patient who is not demented, but who complains of memory problems and exhibits symptoms of memory loss in daily life is familiar to clinicians around the world. Such patients may be reassured that their memory problems are "normal" for their age, or diagnostic terms such as "benign senescent forgetfulness" may be applied to distinguish the condition from dementia. Both of these approaches present certain problems in clinical practice and in research.

Whether the older patient with memory problems ir or is not "normal" depends, of course, on the reference group with whom he is compared. The reference chosen by patients themselves is usually internal. That is, patients complain that their memory abilities have declined from what they once were, and rarely complain that their abilities are worse than the average person 60, 70, or 80 years of age. As in the case of physical abilities, the concern of most 75 year olds is not that they are less robust than their peers, but that they and their peers are less robust than they once were. Thus, the reference group by which older persons generally define problems related to diminished capacities is young, healthy, normal persons. By contrast, the clinician who seeks to assure the older patient that he is "normal" generally has in mind other persons of the same age, many of whom may very well share the same problems.

Diagnostic terms such as "benign senescent forgetfulness" may also be problematic. These terms may be intended to describe clinical conditions in which memory loss is relatively modest and is not marked by a rapidly deteriorating course. However, diagnostic criteria are generally ambiguous and specific criteria may be lacking altogether. Also, the terms may apply to specific subgroups of older individuals with memory loss, not to the typical elderly patient. Finally, as in defining normality, the reference group with whom patients are compared in establishing a diagnosis is generally, at least by implication, other persons of the same age.

Aside from clinical implications, issues related to diagnostic terminology may have important implications for the development of effective treatments for memory impairment in later life. It is noteworthy that age-related memory changes seen in mamallian species other than man have been altered through various pharmacologic treatments (Bartus, Dean, & Beer, 1983). However, all too often, drugs found effective in producing memory changes in healthy aged animals have been evaluated clinically only in neurologically diseased aged adults. The results of such clinical trials have generally been quite disappointing. It is possible, however, that drugs found effective in reversing age-related memory deficits in animals may, indeed, be effective in reducing analagous deficits in humans. Relatively few clinical trials have been undertaken thus far to examine this possibility.

Not only may age-related memory deficits be reversible pharmacologically, but it may be that drugs, or classes of drugs, of possible utility in treating disorders such as Alzheimer's disease (AD) can be identified among healthy aged adults. There is substantial evidence (Gottfries, 1985) that the same neurochemical deficits that underlie age-related memory deficits may also underlie AD. In healthy elderly adults, however, neuronal circuitry may be largely intact and the probability of producing clinical improvement may be greater. Also, the behavioral repertoire of normal aged persons is so much more diverse than that of demented patients, that subtle drug effects may be detected in the former group and overlooked altogether in the latter.

One of the reasons that drug development, particularly in the United States, has not focused on age-related memory impairment is that regulatory authorities have argued that drugs should be targeted at clearly defined, recognized clinical entities, not at vague constellations of symptoms or imprecisely defined clinical conditions (Leber, 1986).

In view of such concerns and the broad clinical and therapeutic implications of issues related to the diagnosis of age-related memory loss, the United States Government's National Institute of Mental Health (NIMH) formed a workgroup of experts to consider diagnostic terminology and criteria. The recommendations of that group have been published (Crook, Bartus, Ferris, Whitehouse, Cohen, and Gershon, 1986) and are now being applied in many international studies, particularly pharmacologic intervention studies.

DIAGNOSTIC TERMINOLOGY: AGE-ASSOCIATED MEMORY IMPAIRMENT

The diagnostic term proposed by the NIMH workgroup was *Age-Associated Memory Impairment* (AAMI). The clinical entity is characterized by complaints of memory impairment in tasks of daily life, substantiated by evidence of such impairment on psychological performance tests with adequate normative data. The performance tests employed should assess recent memory for verbal and nonverbal material. The term AAMI is applied to people over 50 years of age, although this does not imply that such impairment is qualitatively different from that seen (less frequently) in younger adults. The term is also nonspecific with regard to etiology and does not necessarily imply that the disorder is nonprogressive (although deterioration beyond a specified point would exclude individuals from a diagnosis of AAMI).

PROPOSED RESEARCH DIAGNOSTIC CRITERIA

The following inclusion and exclusion criteria were proposed for selecting research participants:

1. Inclusion criteria
   a. Males and females at least 50 years of age.
   b. Complaints of memory loss reflected in such everyday problems as difficulty remembering names of individuals following introduction, misplacing objects, difficulty remembering multiple items to be purchased or multiple tasks to be performed, problems remembering telephone numbers or mailing codes, and difficulty recalling information quickly or following distraction. Onset of memory loss must be described as gradual, without sudden worsening in recent months.
   c. Memory test performance that is a least 1 standard deviation below the mean established for young adults on a standardized

test of secondary memory (recent memory) with adequate normative data. Examples of specific tests and appropriate cutoff scores are listed below, although other measures with adequate normative data are equally appropriate.

Test	Cutoff Score
Benton Visual Retention Test (number correct, Administration A)	7 or less
Logical Memory subtest of the Wechsler Memory Scale (WMS)	6 or less
Associate Learning subtest of the WMS (score on "hard" associates)	6 or less

d. Evidence of adequate intellectual function as determined by a scaled score of at least 9 (raw score of at least 32) on the Vocabulary subtest of the Wechsler Adult Intelligence Scale.

e. Absence of dementia as determined by a score of 24 or higher on the Mini-Mental State Examination (Folstein, Folstein & McHugh, 1975). Many investigators have chosen a score of 27 rather than 24 to exclude questionable cases of dementia.

2. Exclusion criteria

a. Evidence of delirium, confusion, or other disturbances of consciousness.

b. Any neurologic disorder that could produce cognitive deterioration as determined by history, clinical neurological examination, and, if indicated, neuroradiologic examination. Such disorders include AD, Parkinson's disease, stroke, intracranial hemorrhage, local brain lesions including tumors, and normal pressure hydrocephalus.

c. History of any infective or inflammatory brain disease including those of viral, fungal, or syphilitic etiologies.

d. Evidence of significant cerebral vascular pathology as determined by a Hachinski Ischemia Score (modified version; Rosen, Terry, Fuld, Katzman, & Peck, 1980) of 4 or more, or by neuroradiologic examination.

e. History of repeated minor head injury (as in boxing) or single injury resulting in a period of unconsciousness for 1 hour or more.

f. Current psychiatric diagnosis according to DSM-III criteria (American Psychiatric Association, 1980) of depression, mania, or any major psychiatric disorder.

g. Current diagnosis or history of alcoholism or drug dependence.

h. Evidence of depression as determined by a Hamilton Depression Rating Scale (Hamilton, 1967) score of 13 or more.

i. Any medical disorder that could produce cognitive deterioration including renal, respiratory, cardiac, and hepatic disease; diabetes mellitus unless well controlled by diet or oral hypoglycemics; endocrine, metabolic, or hematologic disturbances; and malignancy not in remission for more than 2 years. Determination should be based on complete medical history, clinical examination (including electrocardiogram), and appropriate laboratory tests.

j. Use of any psychotropic drug or any other drug that may significantly affect cognitive function during the month prior to psychometric testing.

TREATMENT STRATEGIES

As noted previously, many of the neurochemcial deficits that underlie AAMI also underlie AD (Gottfries, 1985) and, thus, the same treatment strategies may be appropriate for both disorders. However, it is important to bear in mind that, although the symptoms of AAMI may be quite disturbing to individuals, they are modest in severity when compared with those seen in AD. Thus, drug side-effects that may be tolerable in treating AD may be considered unacceptable in treating AAMI.

A number of diverse and creative strategies have been developed to guide the developed to guide the development of effective treatments for AD and AAMI (see, for example, Crook, Bartus, Ferris, & Gershon, 1986). The following paragraphs focus largely on classes of compounds in which data from clinical trials have been published. Omitted, however, is discussion of compounds such as vasodilators tested in the past and found generally ineffective (Crook, 1985a,b).

Cholinergic Compounds

The cholinergic hypothesis of cognitive dysfunction has passed into its second decade and, in general, attempts at treatment that are based on the hypothesis have been disappointing. Following reports in 1976 by Davies and Maloney and by Bowen and his colleagues of a depletion of choline acetyltransferase in the brains of AD patients, literally dozens of trials with cholinergic agents were conducted in AD. Generally disappointing results have been reported with acetylcholine precursors (for example, Ferris, Reisberg, Crook, Friedman, Schneck, Mir, Sherman, Corwin, Gershon, & Bartus, 1982); with cholinesterase inhibitors such as physostigmine (Stern, Sano, and Mayeux, 1987); and with muscarinic agonists such as arecoline or RS-86 (Bruno, Mohr, Gillespie, Fedio, & Chase, 1986). Investigators who have reported clinically significant effects in AD are subject to criticism on methodologic grounds (Summers, Majovski, Marsh, Tachiki, & Kling, 1986), or for positive interpretation of essentially negative data (Mohs, Davis, Johns, Mathe, Greenwald, Horvath, & Davis, 1985).

It is important to note, however, that 1) cholinergic deficits occur as a function of normal aging (Bartus, Dean, & Fisher, 1986), 2) that selected cholinergic compounds do produce significant effects on memory in healthy aged animals (Bartus, Dean, Beer, Lippa, 1982), and 3) that trials with appropriate cholinergic agents have not been reported in AAMI. Such trials may not be advisable with currently available cholinesterase inhibitors or muscarinic agonists because of side-effects (Dysken and Janowski, 1985; Caine, 1986), but safer, more selective compounds may merit attention in AAMI.

Compounds Affecting Other Neurotransmitter Systems

One reason for the disappointing therapeutic results with cholinergic compounds in AD is probably that other neurotransmitters are also involved in the disorder. For example, loss of noradrenergic cells in the locus coeruleus has been reported in some AD patients (Bondareff, Mountjoy, & Roth, 1982), as has loss of serotonergic cells in the Raphe nucleus (Mann & Yates, 1983). Dopamine (Yates, Allison, Simpson, Maloney, & Gordon, 1979) and GABA (Rossor, Garrett, Johnson, Mountjoy, Roth, & Iverssen, 1982) may also be implicated in AD, although the case is less clear.

The most reasonable compounds for noradrenergic intervention in AAMI may be alpha-2 agonists such as clonidine or guanfacine (Zornetzer, 1986). Animal studies with these compounds have been encouraging (Arnsten & Goldman-Rakic, 1987), but clinical data is limited largely to studies in Korsakoff's disease (McEntee & Nair, 1980) where clonidine has been shown to improve memory in some patients. Clinical trials with guanfacine are now underway in both AD and in AAMI and clonidine is being tested in AD.

There is reasonable evidence based on animal studies that correcting a serotonergic (5-HT) deficit in AAMI may improve memory (Altman & Normile, 1986). Clinical trials with the 5-HT reuptake inhibitors alaproclate and zimelidine have produced equivocal, generally negative, results (Cutler, Haxby, Kay, Narang, Leska, Costa, Nimos, Linnoila, Potter, & Renfrew, 1985; Dehlin, Hedenrud, Jansson, & Norgard, 1985) in AD but the compounds have not been studied in AAMI. Future trials with 5-HT agonists or reuptake inhibitors would appear to be of interest in AAMI and it may be of interest to examine such compounds in combination with drugs affecting other relevant neurotransmitter systems (Carlsson, 1981).

GABA-agonist therapy in AD was evaluated in a recent, well-controlled trial that produced negative results (Mohr, Bruno, Foster, Gillespie, Cox, Hare, Tamminga, Fedio, & Chase, 1986). Multiple trials with dopaminergic compounds in AD have also resulted in largely negative findings (for example, Fleischbracher, Buchgeher, & Schubert, 1986). Evidence of significant deficits in these systems with age may be less compelling and direct intervention through either of these neurotransmitter systems alone would not appear particularly promising in AAMI.

Alteration of monoaminergic (MAO) neurotransmission may hold some promise in treating AD and AAMI. Increased MAO-B activity has been reported in critical brain regions in AD and in normal aging, and clinical evaluation of MAO-B inhibitors such as deprenyl has been proposed (Carlsson, 1983). Empirical evidence for some effects in AD is provided by a recent study of Tariot and his colleagues (1987) demonstrating cognitive and behavioral improvement in AD patients with deprenyl at a low dosage level (at which only MAO-B is thought to be inhibited) and not at a higher level (at which MAO-A is also thought to be inhibited). This line of clinical inquiry may merit further inquiry in AAMI.

Neuropeptides

Numerous studies demonstrate a decline with age in the brain levels of peptides that affect learning and memory (for a review, *see* Banks & Kastin, 1986). There has also long been a body of evidence demonstrating that administration of various neuropeptides or their analogs can facilitate learning and memory in animals under various experimental conditions (de Wied & Gispen, 1977).

Relatively few clinical trials have been conducted with appropriate neuropeptides in AAMI or comparable populations, and trials conducted in AD have been generally disappointing. Among the compounds tested most extensively in AD is a synthetic ACTH 4-9 analog, Organon 2766, found to possibly affect mood but not cognition (Berger & Tinklenberg, 1981; Soinenen, Koskinen, Helkala, Pigache, & Riekkinen, 1985). Similar results were reported in early studies with ACTH 4-10 (Ferris, Sathananthan, Gershon, Clark, & Machinsky, 1976). Studies have also focused on vasopressin (VP) and various VP analogs, including 1-desamino-8-D-arginine vasopressin (DDAVP) and desglycinamide-9-arginine-8-vasopressin (DGAVP) in the treatment of AD. As in the case of ACTH, small changes in behavioral performance have been repeatedly observed, but these changes have generally been ascribed to changes in attention or mood rather than learning or memory (Peabody, Thiemann, Pigache, Miller, Berger, Yesavage, & Tinklenberg, 1985).

Trials in AD with thyrotropin releasing hormone (TRH) or TRH analogs (such as MK-771) have been suggested (Davies, 1981; Yarbrough & Pomara, 1985) based, in part, on the unique facilatory effects of the peptide on cholinergic neurons. Clinical evidence is extremley limited, but a small, preliminary study of TRH has been reported, with negative results (Peabody, Deblois, & Tinklenberg, 1986). The opiate antagonist naloxone has been shown to influence attention, learning, and memory in different animal and human paradigms (for example, Arnsten, Segal, & Neville, 1983; Gallagher, 1982), but neither the compound, nor its oral analog naltrexone, were found to be clinically effective in AD (Tariot, Sunderland, Weingartner, Murphy, Cohen, & Cohen, 1986).

Although clinical trials conducted to date with neuropeptides in AD have been rather discouraging, trials of selected compounds in AAMI may merit consideration. It should be bourne in mind that the clear facilatory effects of these compounds in animal models of learning and memory may bear more relevance to AAMI than to the treatment of severely impaired AD patients.

"Nootropics"

The term "nootropic" was coined by

Giurgea (1976) to describe compounds that directly affect higher brain function and metabolism and have virtually no physiologic effects at other body sites. Piracetam, the prototypical nootropic, is a GABA analog that has diverse effects on brain chemistry and facilitates performance on various learning and memory paradigms in both humans and animals (Giurgea, 1976). A number of trials have been conducted with piracetam in AD and, in general, results have been equivocal or negative (Ferris, Reisberg, Crook, Friedman, Schneck, Sherman, Corwin, Gershon, & Bartus, 1982). However, carefully controlled trials with the compound have not been reported in AAMI. This would seem a reasonable undertaking in view of the diverse, potentially facilitative, neurochemical effects of piracetam and its demonstrated effects in reversing age-related behavioral deficits in animals (Bartus, Dean, Sherman, Friedman, & Beer, 1981).

Various analogs of piracetam have been developed, including pramiracetam, aniracetam, oxiracetam, and others. Compounds such as CI-911 and CI-933, vincamine and its analogs, and other compounds may also be referred to as nootropics although they are not related to piracetam and may affect sites outside the brain. Numerous clinical trials have been conducted with such compounds in diverse patient populations, including AD and related disorders. It appears unlikely from this body of evidence that these compounds are of greater utility than piracetam in AD and that, like piracetam, clinical trials in AAMI may be more appropriate than trials in AD.

In general, the concept of a "nootropic" agent is certainly appealing, and careful clinical trials in this condition would be of interest.

New Treatment Strategies

Among other approaches to treatment that may be considered in AAMI are the following:

1. Compounds that alter membrane phospholipids may hold promise in the treatment of AAMI (Rotrosen, 1986). A formulation of phosphatidylserine from bovine cortex (BC-PS) has been shown to exert clinical effects in AD (Delwaide, Gyselynck-Mambourg, Hurlet, & Ylieff, 1986; SMID group, 1988) and is now being carefully evaluated in both AD and AAMI.

2. As discussed by Sudilovsky elsewhere in this volume, alteration of the brain angiotensin-renin system with compounds such as captopril may exert effects on memory. Studies with captopril are now underway in AAMI.

3. Compounds that affect neurotransmitter release through interaction with calcium homeostasis have been developed and may merit evaluation in AAMI (Gibson, 1986). A preliminary study of 4-aminopyridine in AD was encouraging (Wesseling, Agoston Van Dam, Pasma, DeWit, & Havinga, 1984). 3-4-Diaminopyridine is said to be one thousand times more potent than 4-aminopyridine and less toxic (Gibson, 1986). Thus, this compound appears to merit serious clinical evaluation. Aside from the aminopyridines, the calcium channel blocker nimodipine is being studied in AD and may merit attention in AAMI as well.

4. Lynch and his colleagues (1986) have suggested that calcium-activated brain proteases are causal agents in brain aging and perhaps in AD. These investigators suggest that pharmacologic modulation of the enzyme calpain would be of therapeutic utility in AAMI and AD.

5. Roberts (1986) has developed a complex neurobehavioral model linking cognitive deficits in AD and AAMI to decrements in sex hormone levels generally, and specifically to decreases in dehydroepiandrosterone (DHEA) and dehydroepiandrosterose sulfate (DHEAS). Preliminary trials with DHEA are now underway. Hormonal therapy has also been proposed as a possible therapeutic strategy by Fillet and his colleagues (1986). These investigators reported equivocal findings in a preliminary estrogen trial in women with AD (Fillit, Weintraub, Cholst, Luine, McEwen, Amador, & Zabriskie, 1986).

6. Sapolsky and McEwen (1986) have argued that glucocorticoids may play a role in age-related memory impairments and have speculated that glucocorticoid synthesis inhibitors, such as metyrapone, or glu-

cocorticoid receptor antagonists may have some clinical utility. Clinical trials based on these creative suggestions have not been published.

In addition to these possibilities, a number of creative strategies for treating AAMI have been described elsewhere (Crook, Bartus, Ferris, & Gershon, 1986).

SUMMARY

In general, then, the term Age-Associated Memory Impairment (AAMI) has been adopted to describe healthy persons over 50 years of age who have experienced memory loss since early life, but who are not demented. Specific diagnostic criteria have been proposed for AAMI and these criteria are now being applied in clinical studies, including a number of pharmacologic treatment studies. Many of these latter studies are supported by evidence from preclinical studies suggesting that age-related memory deficits in animals can be pharmacologically altered. This evidence gives reason to hope that an important behavioral deficit associated with aging may be controlled or corrected through drug treatment. The development of such a treatment would be an accomplishment of immense clinical and scientific significance.

RESUMEN

La expresión "Defecto en la memoria asociada a la edad" (Age-Associated Memory Impairment-AAMI) ha sido adoptada para describir el estado de personas sanas mayores de 50 años de edad que han experimentado pérdida de memoria desde temprana edad, pero que no están dementizadas y que no es probable que lleguen a ser dementes. Criterios diagnósticos específicos han sido propuestos para el AAMI y estos criterios están siendo aplicados en estudios clínicos, incluyendo observaciones de tratamiento farmacológico. Muchos de estos últimos estudios son apoyados por la evidencia de estudios preclínicos sugestivos de que defectos en la memoria relacionadas con la edad en animales, pueden ser modificados por fármacos.

Esta evidencia sostiene la esperanza de que un importante defecto en la conducta asociado con el envejecimiento puede ser controlado o corregido por tratamiento medicamentoso. El desarrollo de este tratamiento será la realización de una conquista clínica y científica de muy alta significación.

RÉSUMÉ

Le terme de "détérioration mnésique en rapport avec l'âge" (Age-Associated Memory Impairment - AAMI) a été généralement adopté pour définir des individus de plus de 50 ans, en bonne santé, qui ont présenté des troubles de la mémoire dès le jeune âge, mais qui ne sont pas déments et dont il est peu probable qu'il le deviendront. Des critères diagnostiques spécifiques ont été proposés qui commencent à être appliqués dans les études cliniques et dans certaines études pharmacologiques à visée thérapeutique. Beaucoup de celles-ci sont dictées par les résultats d'études précliniques suggérant que chez l'animal, de tels troubles de la mémoire peuvent être pharmacologiquement modifiés. Il y a là une raison d'espérer qu'un important déficit comportemental en relation avec le vieillissement peut être controlé ou corrigé par un traitement médicamentex. La mise au point d'un tel traitement serait un projet d'une immense signification clinique et scientifique.

ZUSAMMENFASSUNG

Der Ausdruck "Gedächtnismangel entsprechend dem Alter" (Age-Associated Memory Impairment-AAMI) wurde angewendet um den Zustand gesunder Personen zu benennen, die, älter als 50 Jahre, einen Gedächtnisverlust in jungen Jahren erfahren

haben, die aber nicht dement geworden sind und bei denen es nicht wahrscheinlich ist, dass sie dement werden. Verschiedene diagnostische Gesichtspunkte sind für den AAMI vorgeschlagen worden und diese Gesichtspunkte werden bei Klinischen Untersuchungen verwendet einschliesslich bei Beobachtungen pharmakologischer Behandlung. Viele dieser letzteren Studien werden unterstützt durqhdie Augenscheinlichkeit sugestiver, vorklinischer Studien, dass Gedächtnismängel in Verbindung mit dem Alter bei Tieren durch Pharmaka geändert werden können.

Diese Offenkundigkeit unterstützt die Hoffnung, dass auch e in bedeutender Verhaltensdefekt in Verbindung mit dem Altwerden kontrolliert oder korrigiert werden kann durch die medikamentöse Behandlung Die Entwicklung dieser Behandlung wird die Verwirklichung einer bedeutenden klinischen und wissenschaftlichen Errungenschaft darstellen.

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Predictive Regulation of Associative Learning in a Neural Network by Reinforcement and Attentive Feedback

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A real-time neural network model is described in which reinforcement helps to focus attention upon and organize learning of those environmental events and contingencies that have predicted behavioral success in the past. Computer simulations of the model reproduce properties of attentional blocking, inverted-U in learning as a function of interstimulus interval, primary and secondary excitatory and inhibitory conditioning, anticipatory conditioned responses, attentional focussing by conditioned motivational feedback, and limited capacity short term memory processing. Qualitative explanations are offered of why conditioned responses extinguish when a conditioned excitor is presented alone, but do not extinguish when a conditioned inhibitor is presented alone. These explanations invoke associative learning between sensory representations and drive, or emotional, representations (in the form of conditioned reinforcer and incentive motivational learning), between sensory representations and learned expectations of future sensory events, and between sensory representations and learned motor commands. Drive representations are organized in opponent positive and negative pairs (e.g., fear and relief), linked together by recurrent gated dipole, or READ, circuits. Cognitive modulation of conditioning is regulated by adaptive resonance theory, or ART, circuits which control the learning and matching of expectations, and the match-contingent reset of sensory short term memory. Dendritic spines are invoked to dissociate read-in and read-out of associative learning and to thereby design a memory which does not passively decay, does not saturate, and can

be actively extinguished by opponent interactions.

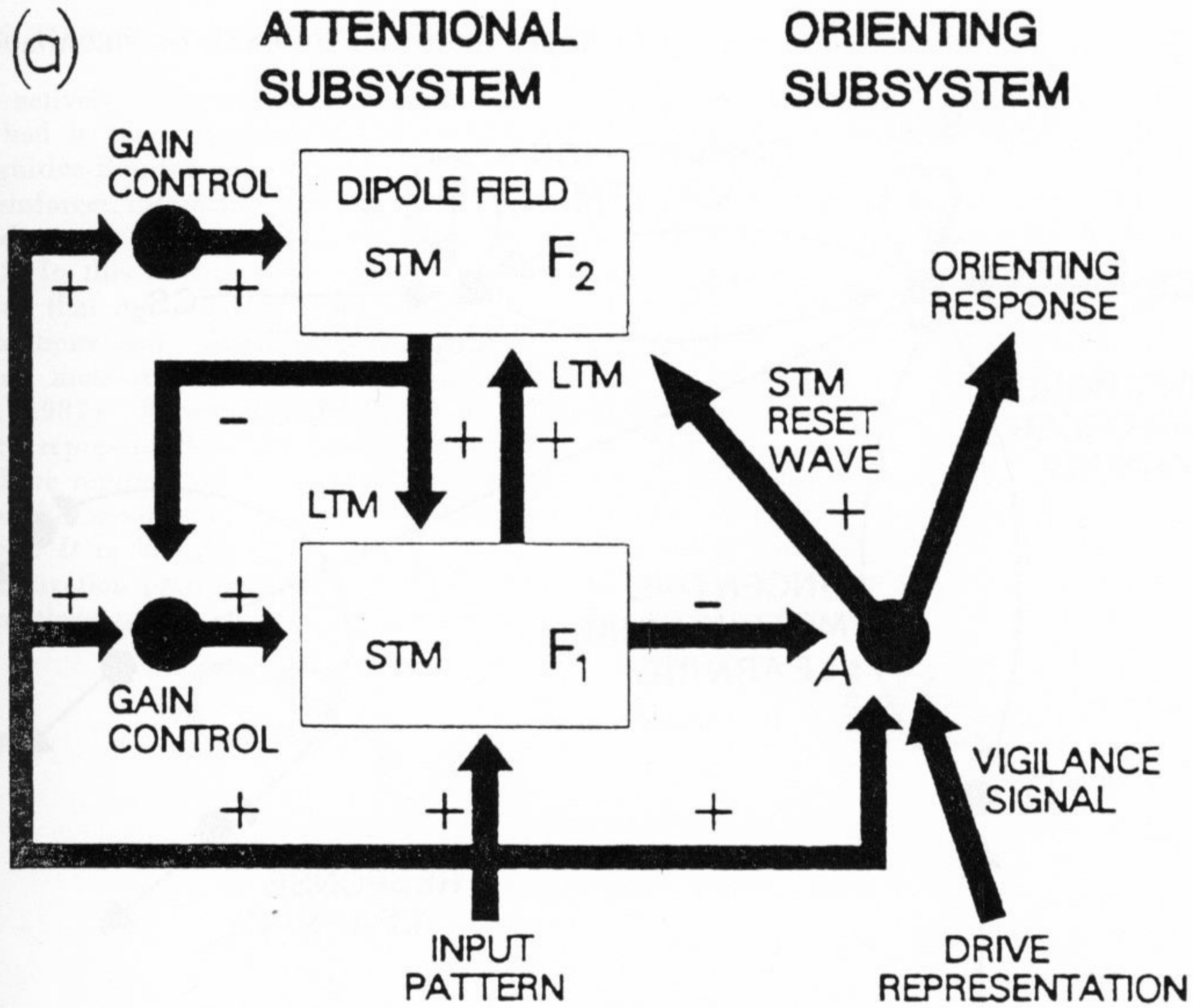
1. Introduction

A key problem in biological theories of intelligence concerns the manner in which external events interact with internal organismic requirements to trigger learning processes capable of focussing attention upon motivationally desired goals. The results reported herein further develop a neural theory of learning and memory (Grossberg, 1982, 1987) in which sensory-cognitive-reinforcement circuits help to focus attention upon and organize learning of those environmental events that predict behavioral sucess.

The first set of results (Grossberg and Levine, 1987) describe computer simulations that show how model reproduces properties of attentional blocking, inverted-U in learning as a function of interstimulus interval, anticipatory conditioned responses, secondary reinforcement, attentional focussing by conditioned motivational feedback, and limited capacity short-term memory processing. Conditioning occurs from sensory to drive representations ("conditioned reinforcer" learning), from drive to sensory representations ("incentive motivational" learning), and from sensory to motor representations ("habit" learning). The conditionable pathways contain long-term memory traces that obey a non-Hebbian associative law. The neural model embodies a solution of two key design problems of conditioning, the synchronization and persistence problems. This model of vertebrate learning has also been compared with data and models of invertebrate learning. Predictions derived from models of vertebrate learning have been compared with data about invertebrate learning, including data from *Aplysia* about facilitator neurons and data from *Hermissenda* about voltage-dependent Ca^{++} currents.

In the second set of results (Grossberg and Schmajuk, 1987), representations are expanded to include positive and negative opponent drive representations, as in the opponency between fear and relief. This expanded real-time neural network model is developed to explain data about the acquisition and extinction of conditioned excitors and inhibitors. Systematic computer simulations have been performed to characterize a READ circuit, which joins together a mechanism of associative learning with an opponent processing circuit, called a *recurrent gated dipole.* READ circuit properties clarify how positive and negative reinforcers are learned and extinguished during primary and secondary conditioning. Habituating chemical transmitters within a gated dipole determine an affective adaptation level, or context, against which later events are evaluated. Neutral CS's can become reinforcers by being associated either with direct activations or with antagonistic rebounds within a previously habituated dipole. Neural mechanisms are characterized whereby conditioning can be actively extinguished, by a process called *opponent extinction,* even if no passive memory decay occurs.

READ circuit mechanisms are joined to mechanisms for associative learning of incentive motivation; for activating and storing internal representations of sensory cues in a limited capacity short term memory (STM); for learning, matching, and mismatching sensory expectancies, learning to the enhancement or updating of STM; and for shifting the focus of attention toward sensory representations whose reinforcement history is consistent with momentary appetitive requirements. This architecture has been used to explain conditioning and extinction of a conditioned excitor; conditioning and extinction of a conditioned inhibitor; properties of conditioned inhibition as a "slave" process and as a "comparator" process, including effects of pretest deflation or inflation of the conditioning context, of familiar or novel training or test contexts, of weak or strong shocks, and of preconditioning US-alone exposures. The same mechanisms have also been used (Grossberg, 1982, 1987) to explain phenomena such as unblocking, overshadowing, latent inhibition, superconditioning, partial reinforcement acquisition effect, learned helplessness, and vicious-circle behavior. The theory clarifies why alterna-



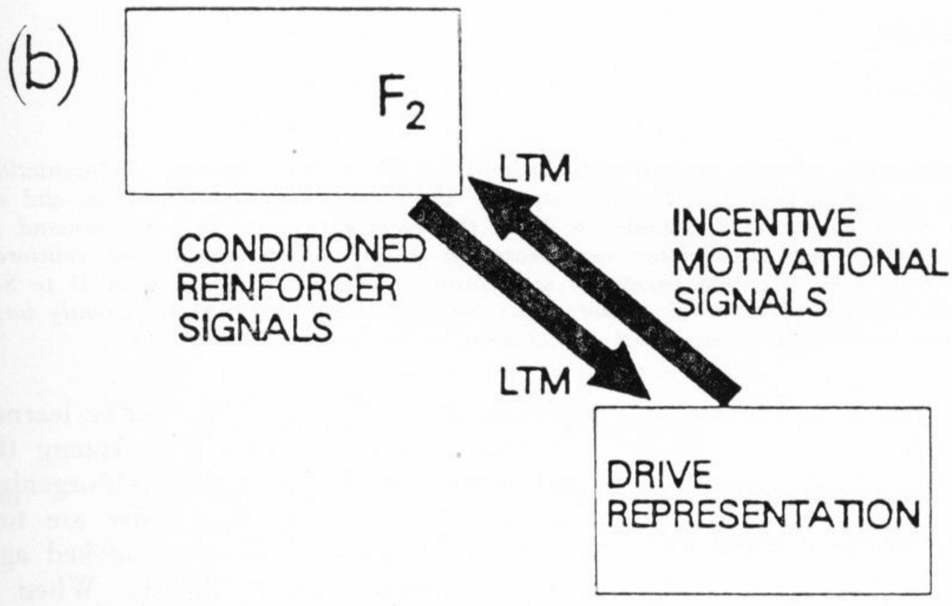


Fig. 1. — *Anatomy of an adaptive resonance theory (ART) circuits (a) Interactions between the attentional and orienting subsystems. Code learning takes place at the long term memory (LTM) traces within the bottom-up and top-down pathways between levels* F_1 *and* F_2. *The top-down pathways can read-out learned expectations, or templates, that are matched against bottom-up input patterns at* F_1. *Mismatches activate the orienting subsystem A, thereby resetting short term memory (STM) at* F_2 *and initiating search for another recognition code. Subsystem* A *can also activate an orienting response. Sensitivity to mismatch at* F_1 *is modulated by vigilance signals from drive representations. (b) Trainable pathways exist between level* F_2 *and the drive representations. Learning from* F_2 *to a drive representation endows a recognition category with conditioned reinforcer properties. Learning from a drive representation to* F_2 *associates the drive representation with a set of motivationally compatible categories. (Adapted from Carpenter and Grossberg, 1987c).*

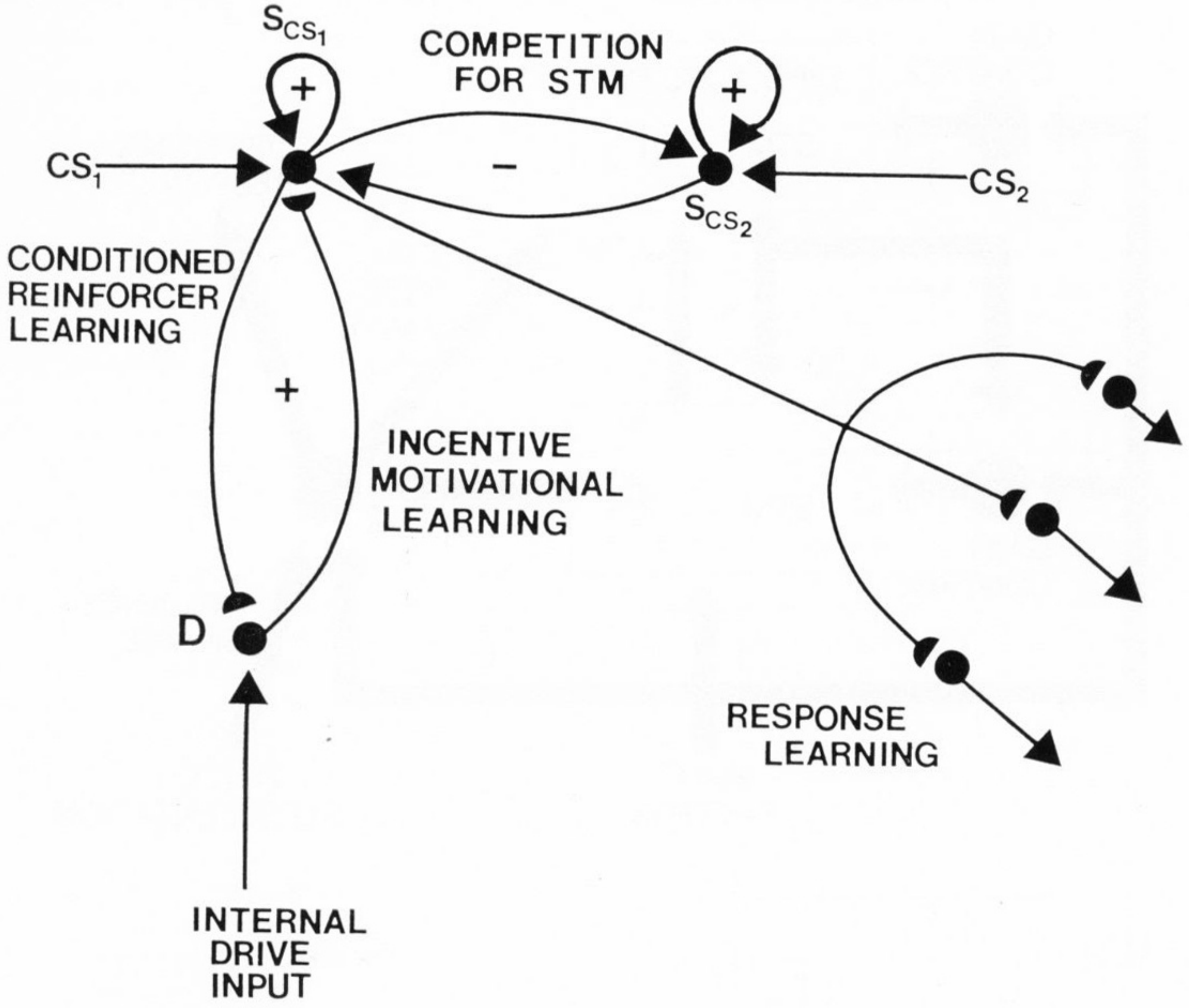


Fig. 2. — *Schematic conditioning circuit: Conditioned stimuli* (CSi) *activate sensory representations* (Scsi) *which compete among themselves for limited capacity short term memory activation and storage. The activated* Scsi *elicit conditioned signals to drive representations and motor command representations. Learning from an* Scsi *to a drive representation* D *is called conditioned reinforcer learning. Learning from* D *to* Scsi *is called incentive motivational learning. Signals from* D *to* Scsi *are elicited when the combination of external sensory plus internal drive inputs is sufficiently large. In the simulations reported herein, the drive level is assumed to be large and constant.*

tive models have been unable to explain an equally large data base.

2. Neural Network Macrocircuits

Two types of macrocircuits control learning within the model.

Sensory-Cognitive Circuit: Sensory-cognitive interactions in the theory are carried out by an Adaptive Resonance Theory (ART) circuit (Carpenter and Grossberg, 1985, 1987a, 1987b; Grossberg, 1976, 1987). The ART architecture suggests how internal representations of sensory events, including conditioned stimuli (CS) and unconditioned stimuli (US), can be learned in stable fashion (Figure 1). Among the mechanisms used for stable self-organization of sensory recognition codes are top-down expectations which are matched against bottom-up sensory signals. When a mismatch occurs, an arousal burst acts to reset the sensory representation of all cues that are currently being stored in STM. In particular, representations with high STM activation tend to become less active, representations with low STM activation tend to become more active, and the novel event which caused the mismatch tends to be more actively stored than it would have been had it been expected.

Cognitive-Reinforcement Circuit: Cognitive-reinforcer interactions in the theory are carried out in the circuit described in Figure 2. In this circuit, there exist cell populations that are separate from sensory representations and related to particular drives and motivational variables (Grossberg, 1972, 1987). Repeated pairing of a CS sensory representation, S_{CS}, with activation of a drive representation, D, by a reinforcer causes the modifiable synapses connecting S_{CS} with D to become strengthened. Incentive motivation pathways from the drive representations to the sensory representations are also assumed to be conditionable. These $S \longrightarrow D \longrightarrow S$ feedback pathways shift the attentional focus to the set of previously reinforced, motivationally compatible cues (Figure 2). This shift of attention occurs because the sensory representations, which emit conditioned reinforcer signals and receive incentive motivation signals, compete, among themselves for a limited capacity short-term memory (STM) via a shunting on-center off-surround anatomy. When incentive motivational feedback signals are received at the sensory representational field, these signals can bias the competition for STM activity towards motivationally salient cues.

1. $$CS_1 \;—\; US$$

$$CS_1 \longrightarrow CR$$

2. $$CS_1 + CS_2 \;—\; US$$

$$CS_2 \not\longrightarrow CR$$

Fig. 3. — *A blocking paradigm. The two stages of the experiment are discussed in the text.*

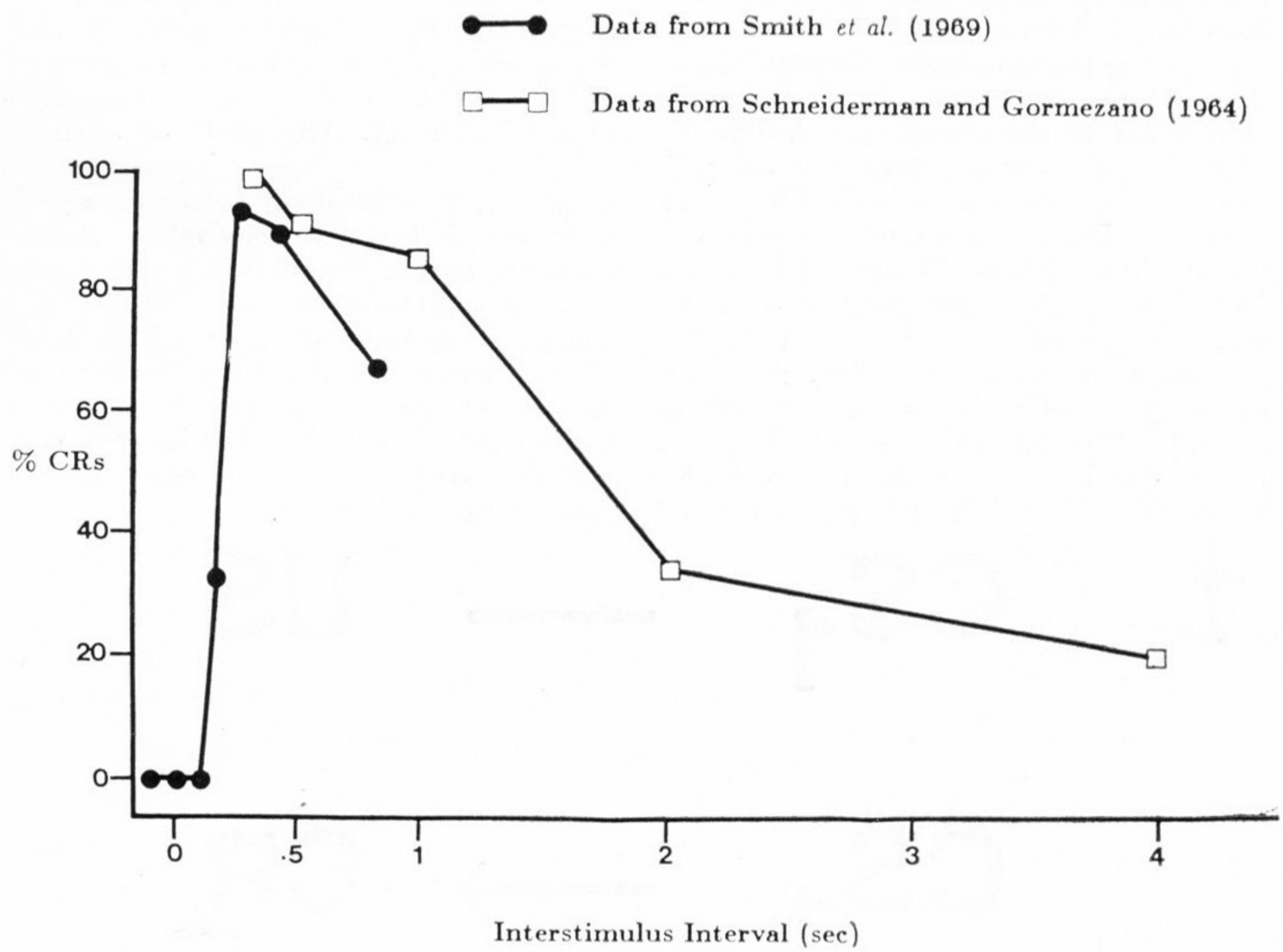


Fig. 4. — *Experimental relationship between conditioned response strength (measured by percentage of trials on which response occurs) and interstimulus interval in the rabbit nictitating membrane response. (Reprinted with permission from Sutton and Barto, 1981).*

3. Attentional Blocking and Interstimulus Interval

The attentional modulation of Pavlovian conditioning is part of the general problem of how an information processing system can selectively process those environmental inputs that are most important to the current goals of the system. A key example is the blocking paradigm studied by Kamin (1969) (Figure 3). First, a stimulus CS_1, such as a tone, is presented several times, followed at a given time interval by an unconditioned stimulus US, such as electric shock, until a conditioned response, such as fear, develops. Then CS_1 and another stimulus CS_2, such as a light, are presented together, followed at the same time interval by the US. Finally, CS_2 is presented alone, not followed by a US, and no conditioned response occurs.

The blocking paradigm suggests four key subproblems of the selective information processing problem. These subproblems are: (1) How does the pairing of CS_1 with US in the first phase of the blocking experiment endow the CS_1 cue with properties of a conditioned, or secondary, reinforcer? (2) How do the reinforcing properties of a cue shift the focus of attention towards its own processing? (3) How does the limited capacity of attentional resources arise, so that a shift of attention towards one set of cues can prevent other cues from being attended? (4) How does withdrawal of attention from a cue prevent that cue from entering into new conditioned relationships?

The explanation of blocking also leads to an explanation of the inverted-U relationship between strength of the conditioned response (measured in one of several ways) and the time interval (ISI) between conditioned and unconditioned stimuli. Figure 4 gives an example of experimental data on the effects of ISI from studies of Smith *et al.* (1969) and Schneiderman and Gormenzano (1964) of the rabbit nictitating membrane response. This is noteworthy because Sutton and Barto (1981) previously stated that the ISI data pose a difficulty for any network with associative synapses, that is, synapses whose efficacy changes as a function of the correlation between presynaptic and postsynaptic activities. They argued that a network with associative synapses should, to a first approximation, have an optimal ISI of zero because cross-correlation between two stimulus traces is strongest when the two stimuli occur simultaneously. To avoid this difficulty, other modellers introduced a delay in the CS pathway that was equal to the optimal ISI. But such a delay would delay the CR by an equal amount, and hence is incompatible with the so-called anticipatory CR that occurs before US onset. On this basis, Sutton and Barto suggested a different synaptic modification rule at the single-unit level.

Our simulations, by contrast, reproduce both the ISI data and the anticipatory CR without invoking a long delay in the CS pathway. Poor conditioning with CS and US simultaneous, or nearly so, is explained by a mechanism identical to the blocking mechanism except that CS_1 is replaced by US and CS_2 by CS. In both cases, the stimulus with more motivational significance inhibits the processing of the stimulus with less motivational significance. Poor conditioning with CS and US far apart in time occurs because by the time the US arrives,

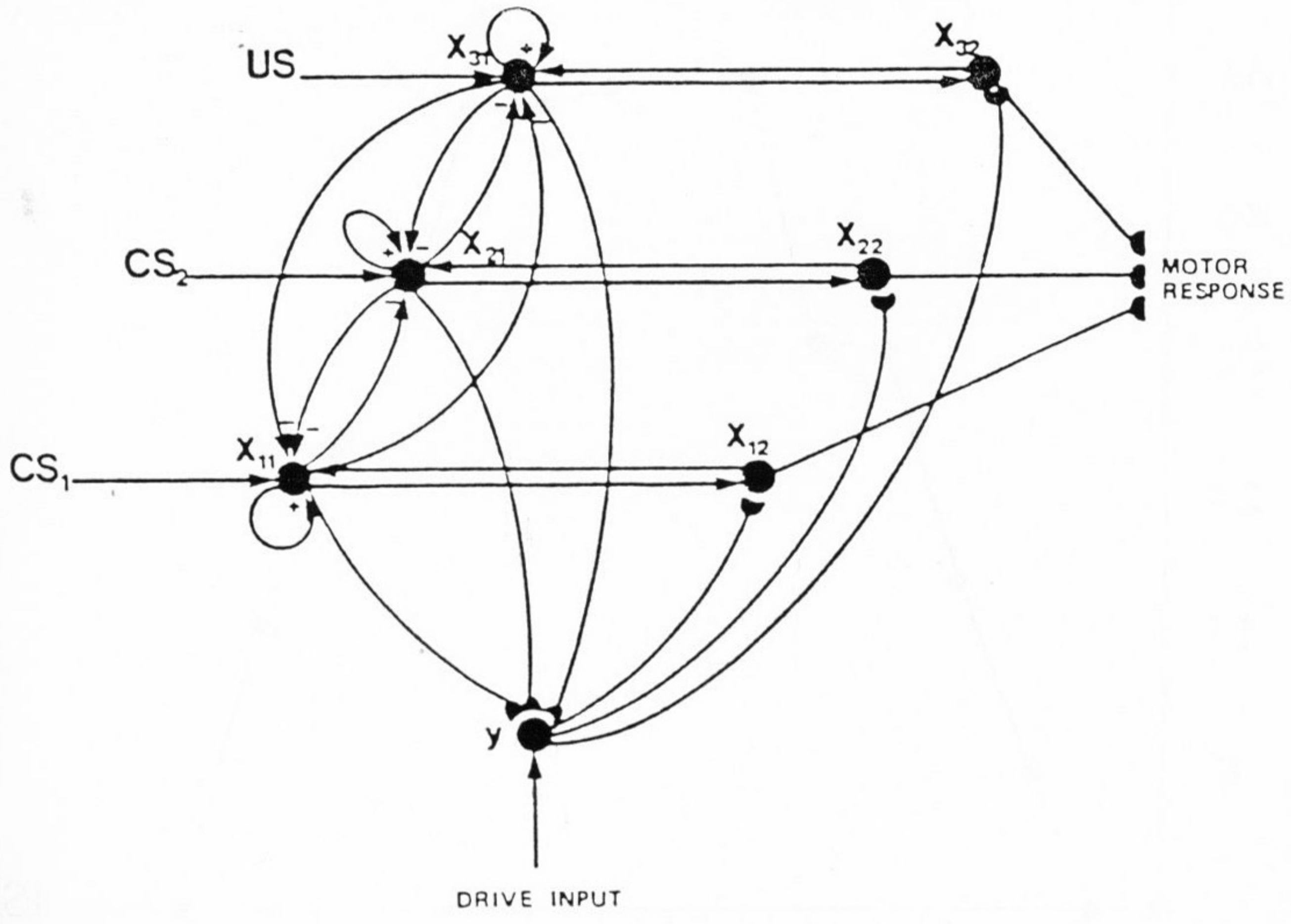


Fig. 5. — *Simulated network: Each sensory representation possesses two stages with STM activities* xi_1 *and* xi_2. *A CS or US input activates its corresponding* xi_1. *Activation of* xi_1 *elicits unconditionable signals to* xi_2 *and conditioned reinforcer signals to* D, *whose activity is denoted by* y. *Incentive motivational feedback signals from* D *activate the second stage potentials* xi_2, *which then send feedback signals to* xi_1. *Conditionable longterm memory traces are designated by hemi-disks.*

the CS representation has decayed in short-term memory to a level that is below the threshold for affecting efficacy of the appropriate synapses.

The answers to subproblems (1) to (4) are obtained from study of a network which includes modifiable associative links between sensory and drive representations (in both directions) and competitive links between different sensory representations (Figure 2). The associative links do not obey Hebb's postulate because cross-correlation is counteracted by decays; hence, synaptic efficacy can either increase or decrease with paired presynaptic and postsynaptic activities (Grossberg, 1968, 1969, 1982), not just increase, as Hebb claimed (Hebb, 1949). Such an associative law has recently received direct neurophysiological support (Levy, Brassel, and Moore, 1983; Levy and Desmond, 1985; Rauschecker and Singer, 1979; Singer, 1983). The existence of drive representations was derived from an analysis of the *synchronization problem* (Grossberg, 1971); that is, of how a stable conditioned response can develop even if variable time lags occur between the CS and the US. These drive representations, separate from the sensory representations of particular stimuli, are what Bower has called *emotion nodes* (Bower, 1981; Bower, Gilligan, and Monteiro, 1981) and Barto, Sutton, and Anderson (1983) have called *adaptive critic elements*. A US unconditionally activates its drive representation if the drive level is sufficiently high. Repeated pairing of a CS with, for example, a food US causes pairing of stimulation of

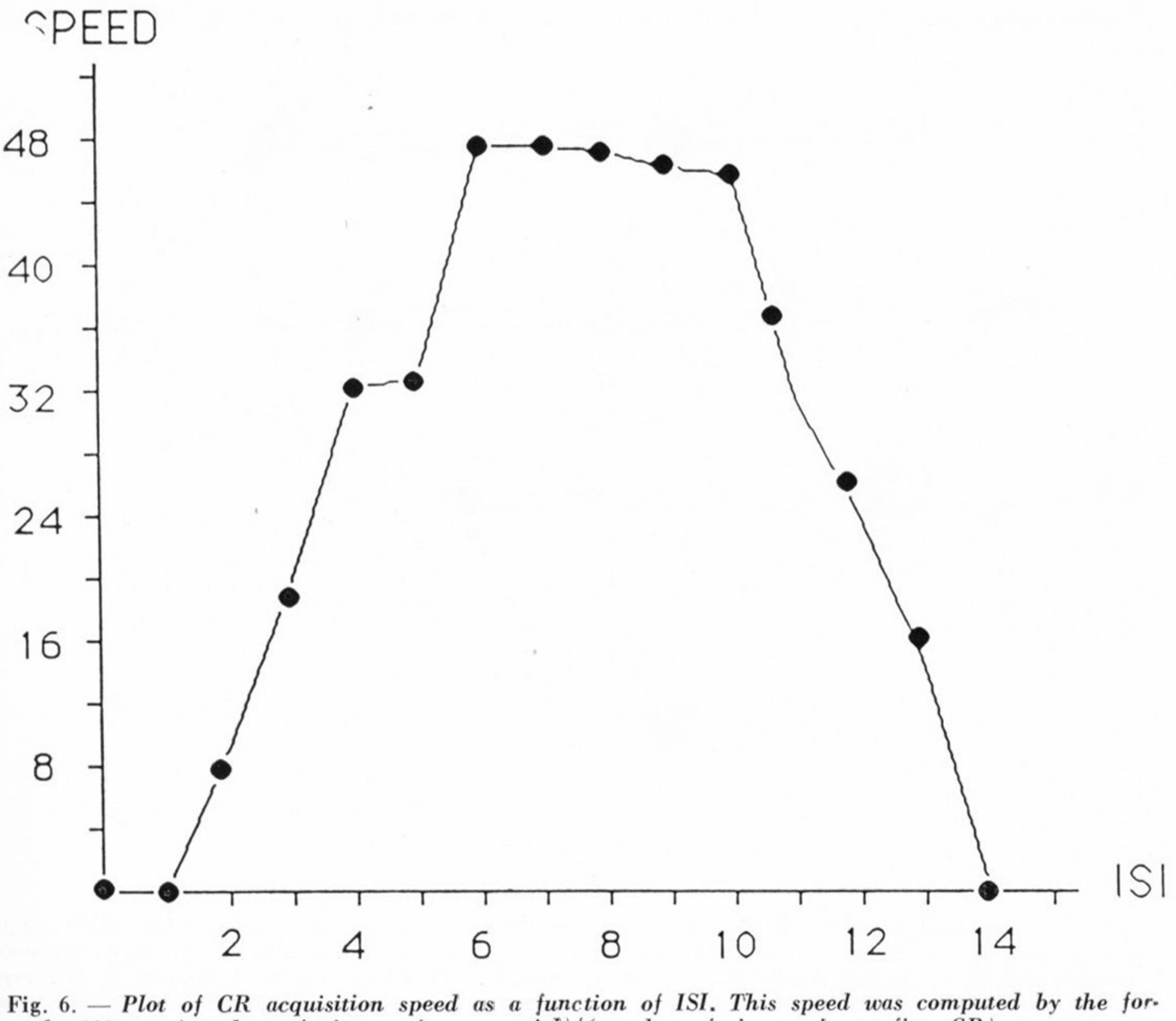
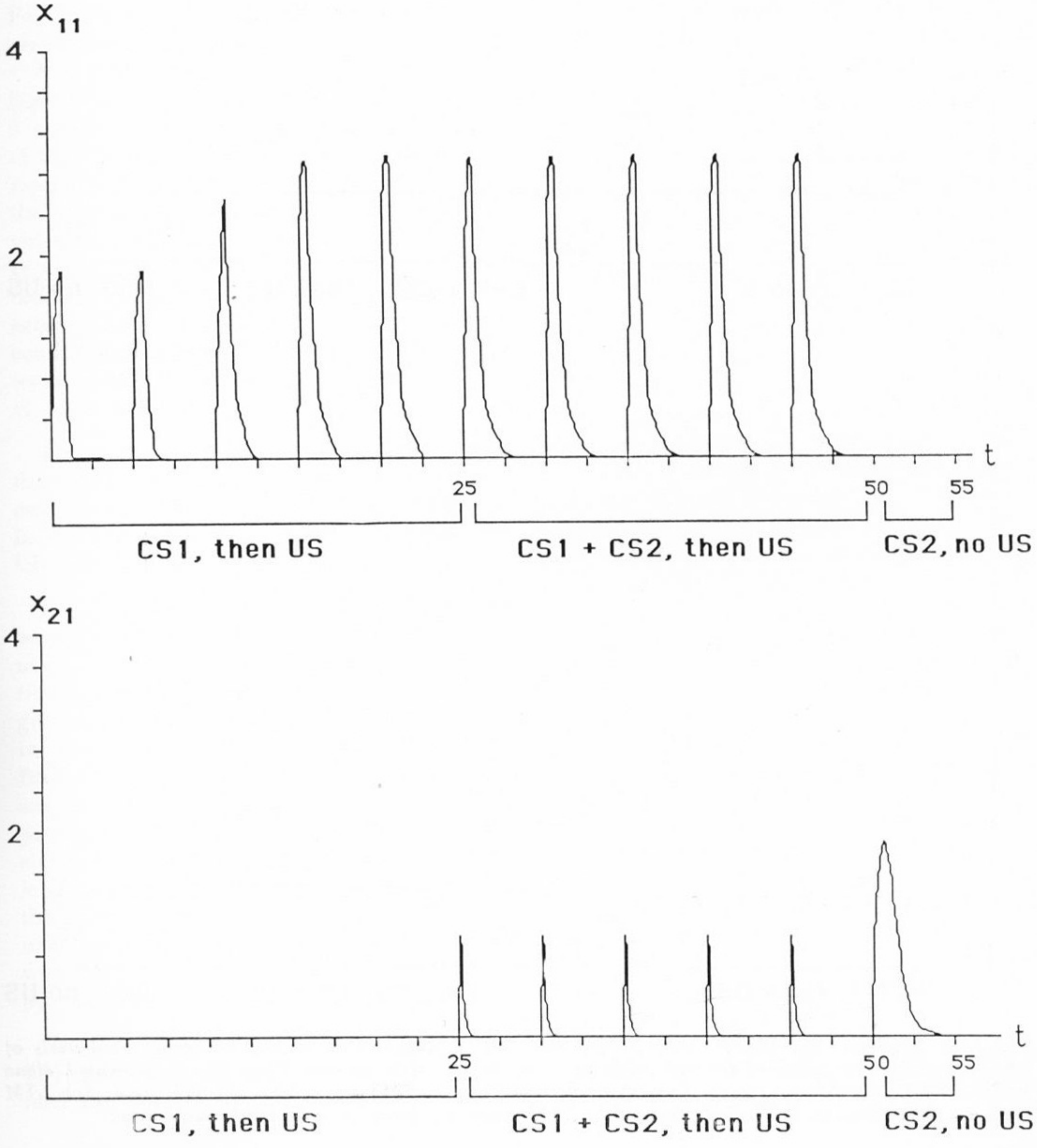


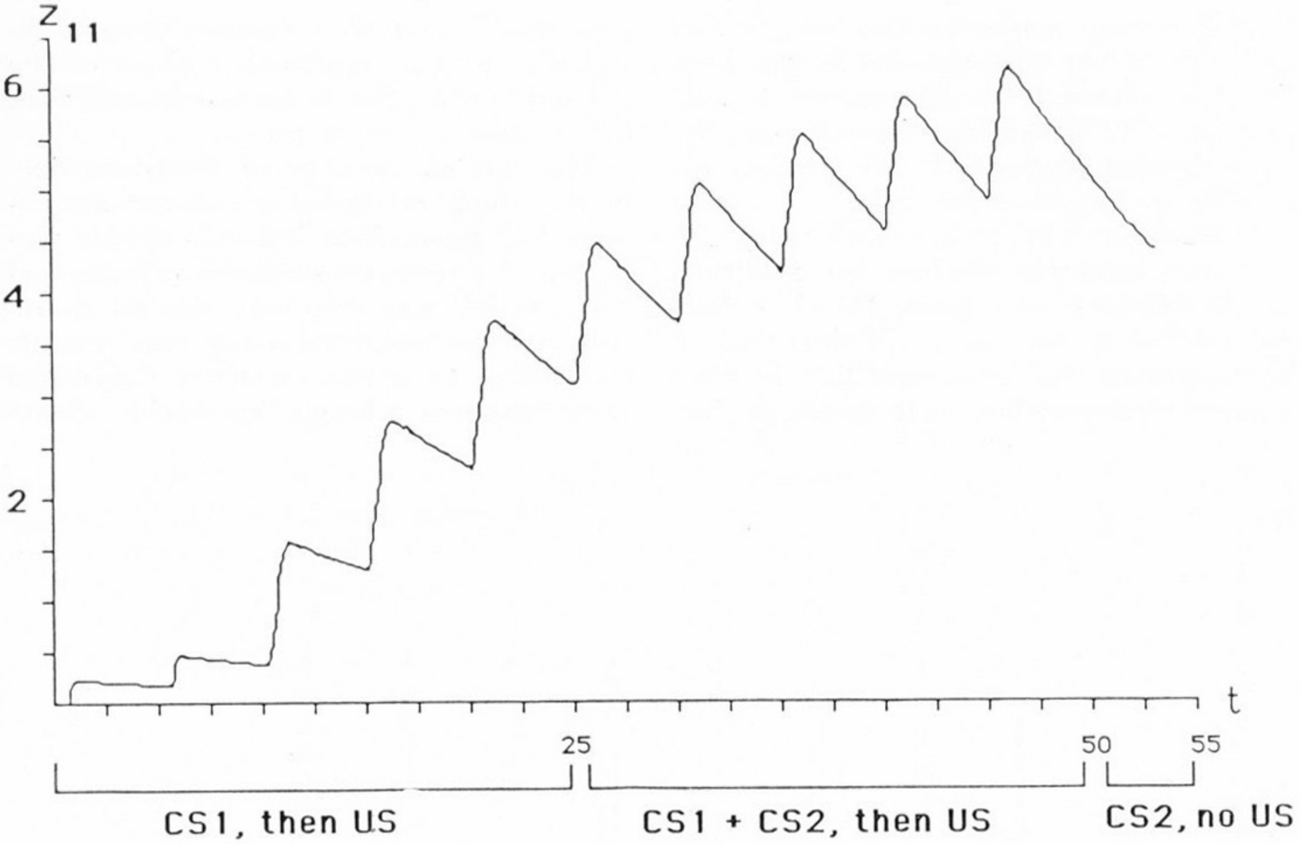
Fig. 6. — *Plot of CR acquisition speed as a function of ISI. This speed was computed by the formula 100 × (number of time units per trial)/(number of time units to first CR).*

the CS sensory representation, denoted *Scs*, with that of the representation for the hunger drive, denoted *Dh*. The answer to subproblem (1) therefore depends on the strengthening of *Scs* $\longrightarrow$ *Dh* synapses according to an associative rule.

Subproblem (2) is answered using *Dh* $\longrightarrow$ *Scs* incentive motivational feedback. In the blocking experment, Scs_1 is enhanced relative to Scs_2. Scs_2 will thus tend to be suppressed due to competition between sensory representations that causes limited capacity of short term memory storage. Similarly, in the simultaneous phase of the ISI experiment, *Sus* in more enhanced than *Scs*, so that *Scs* is suppressed.

The limited capacity of short-term memory, which is needed to answer subproblem (3) arises from limited capacity properties of a recurrent on-center off-surround field, which was originally derived to satisfy a more basic processing requirement: the ability to process spatially distributed input patterns without irreparably distor-





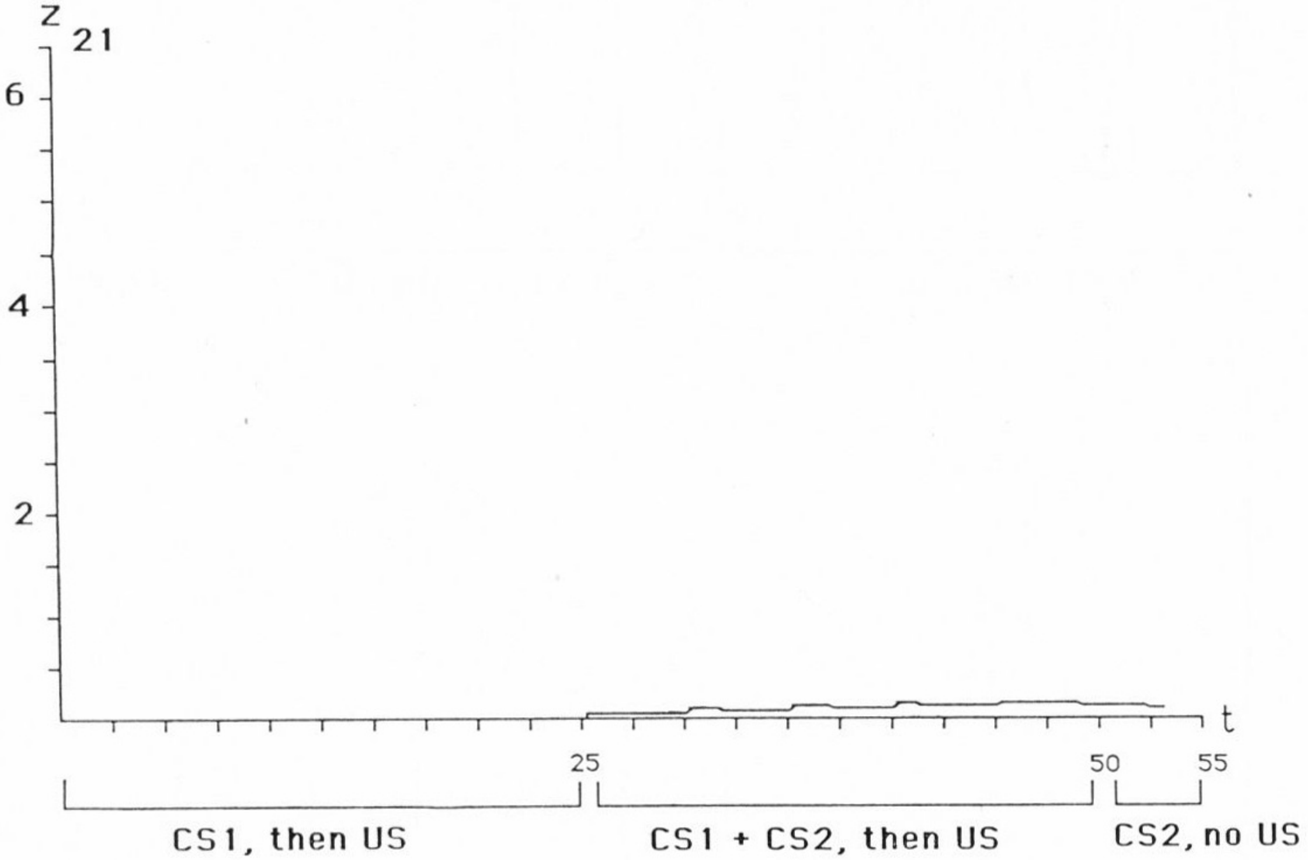


Fig. 7. — *Blocking simulation: In (a)-(d), the ISI = 6 between* CS_1 *and US onset. Five trials of* CS_1*-US pairing are followed by five trials of* $(CS_1 + CS_2)$*-US pairing. Then* CS_2 *is presented alone for one trial. (a) Activity* x_{11} *of* Ccs_1 *through time; (b) Activity* x_{21} *of* Scs_2 *through time; (c) LTM trace* z_{11} *from* Scs_1 *to* D *through time; (d) LTM trace* z_{21} *from* Scs_2 *to* D *through time.*

ting these patterns due to either noise or saturation (Ellias and Grossberg, 1975; Grossberg and Levine, 1975). Figure 2 schematizes a network with modifiable sensory-to-drive and drive-to-sensory association links and recurrent on-center off-surround links between sensory representations.

Our computer simulations, reported more completely in Grossberg and Levine (1987), run through different stimulus conditions on the network of Figure 5, which is a variant of Figure 2 with three sensory representations, CS_1, CS_2, and US. For simplicity, there is only one drive representation, D, in our network. The $US \longrightarrow D$ and $D \longrightarrow US$ synapses are fixed at high value. The $CS \longrightarrow D$ and $D \longrightarrow CS$ synapses are strengthened by appearance of the US while the CS short term memory representation is active. In this variant of the network, sensory representations are divided into two successive stages. The activity xi_1 of the ith first stage can activate conditioned reinforcer pathways, whereas the activity xi_2 of the ith second stage receives conditioned incentive motivational pathways from D, and can thereupon activate xi_1 and output motor pathways.

The same set of network parameters yielded both the ISI inverted-U curve in the case of only one CS present, and blocking in the case of two CS's. In both cases, the CR anticipated the US.

Our simulated ISI curves (Figure 6) were qualitatively compatible with experimental data on the rabbit's conditioned nictitating membrane response shown in Figure 4. For ISI's of fewer than 2 time units in the numerical algorithm, competition from the US representation prevented CS activity from staying above the $Scs \longrightarrow D$ pathway's threshold long enough to appreciably increase the pathway's strength while D was activated by the US. At long ISI's, the prior decay of the CS's short term memory trace prevented the $Scs \longrightarrow D$ pathway from sensing the later activation of D by the US.

In the blocking simulation (Figures 7a-7d), pairing of CS_1 with a delayed US enabled the long term memory trace of the $CS_1 \longrightarrow D$ pathway to achieve an S-shaped cumulative learning curve. After CS_1 had become a conditioned reinforcer, it enhanced its own short term memory storage by generating a large $Scs_1 \longrightarrow D \longrightarrow Scs_1$ feedback signal. As a result, when CS_1 and CS_2 were simultaneously presented, the short term memory activity of Scs_2 was quickly suppressed by competition from CS_1. Consequently, the long term memory $Scs_2 \longrightarrow D$ pathway did not grow in strength, preventing the CS_2 from being a conditioned reinforcer or eliciting a CR.

4. Comparison with Aplysia Conditioning Model

An alternative explanation of blocking, due to Hawkins and Kandel (1984), involved habituation of transmitter pathways. Based on invertebrate evidence, they developed a model whereby each US activates a *facilitator neuron* that presynaptically modulates CS pathways. They explain blocking (p. 385) by saying that "the output of the facilitator neurons decreases when they are stimulated continuously". Thus after a CS_1 is paired with a US on a number of trials, subsequent presentation of a compound stimulus $CS_1 + CS_2$ with a US does not condition CS_2 because the facilitator neuron cannot fire adequately. Hawkins and Kandel's explanation, however, is incompatible with the fact (Kamin, 1969) that blocking can be overcome ("unblocked") if $CS_1 + CS_2$ is paired with either a higher or lower intensity of shock than CS_1 alone. Recent evidence (Matzel *et al.* 1985) indicates that unblocking can also occur if the response to CS_1 is extinguished.

In our framework, the explanation for unblocking depends on *gated dipole* opponent processes that link together "positive" and "negative" drive representations (Figure 8). Positive and negative channels allow for a comparison between current and expected levels of positive or negative reinforcement. The more complete theory of Grossberg (1982, 1987) which includes gated dipoles has explained such unblocking results quantitatively.

In the remainder of the article, some of our computer simulation results using gated

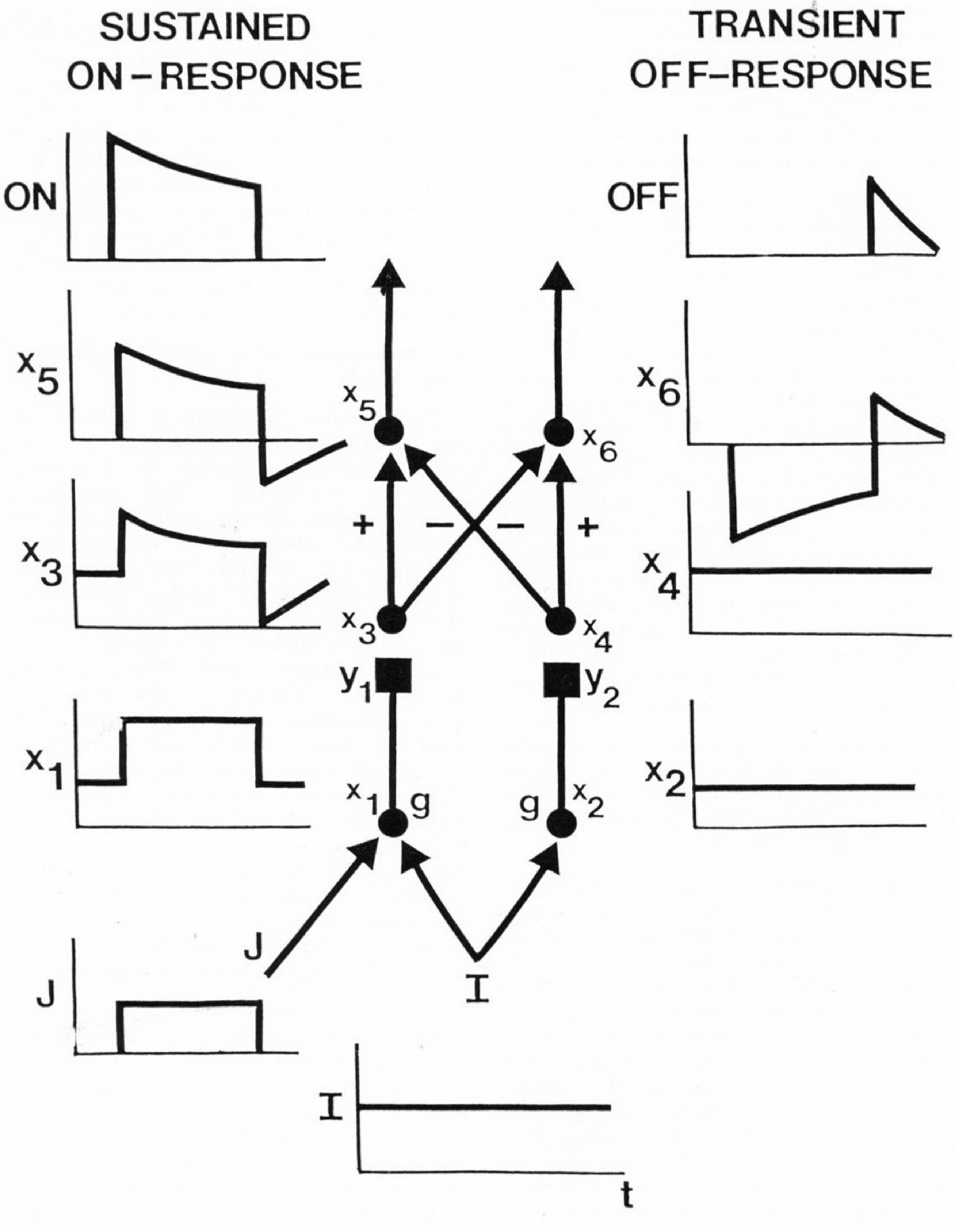


Fig. 8. — *Example of a feedforward gated dipoles A sustained habituating on-response (top left) and a transient off-rebound (top right) are elicited in response to onset and offset, respectively, of a phasic input* J *(bottom left) when tonic arousal* I *(bottom center) and opponent processing (diagonal pathways) supplement the slow gating actions (square synapses). See text for details.*

dipoles are summarized. A more systematic development is provided in Grossberg and Schmajuk (1987). Such gated dipoles are needed because, in the cognitive-reinforcement circuit, CS's are conditioned to either the onset or the offset of a reinforcer. In order to explain how the offset of a reinforcer can generate an antagonistic rebound to which a simultaneous CS can be conditioned, gated dipoles were introduced by Grossberg (1972). A gated dipole is a minimal neural network which is capable of generating a sustained, but habituative, on-response to onset of a cue, as well as a transient off-response, or antagonistic rebound, to offset of the cue.

5. The READ Circuit: A Synthesis of Opponent Processing and Associative Learning Mechanisms

Although several varieties of a gated di-

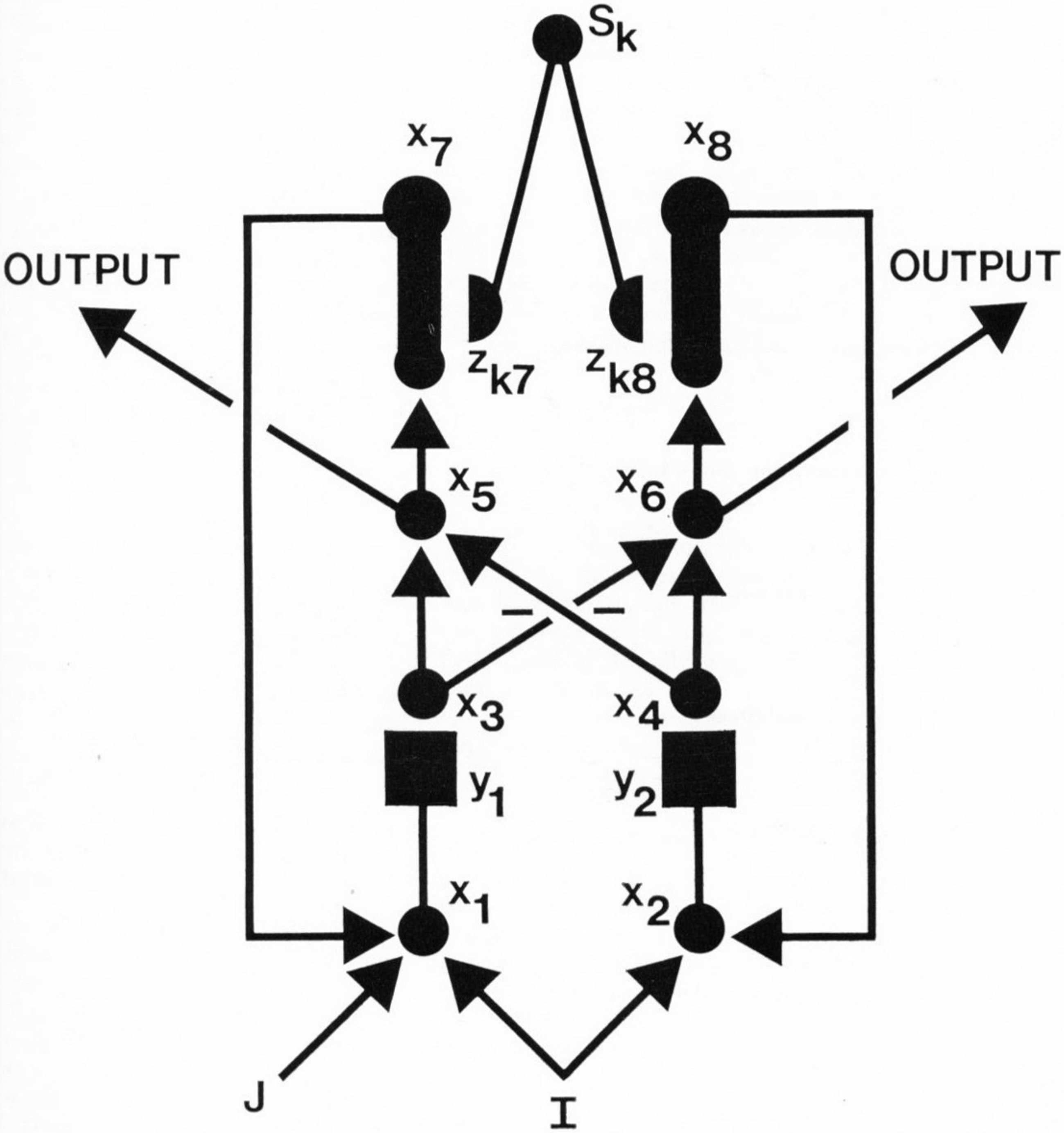


Fig. 9. — *A READ I circuit: This circuit joins together a recurrent gated dipole with an associative learning mechanism. Learning is driven by signals* Sk *from sensory representations* Sk *which activate long term memory (LTM) traces* zk_7 *and* zk_8 *that sample activation levels at the on-channel and off-channel, respectively, of the gate dipole. See text for details.*

The equations for the READ circuit are as follows:

Arousal + US + Feedback On-Activation:

$$\frac{d}{dt}x_1 = -A_1x_1 + I + J + T(x_7) \tag{1}$$

Arousal + Feedback Off-Activation:

$$\frac{d}{dt}x_2 = -A_2x_2 + I + T(x_8) \tag{2}$$

On-Transmitter:

$$\frac{d}{dt}y_1 = B(1 - y_1) - Cg(x_1)y_1 \tag{3}$$

Off-Transmitter:

$$\frac{d}{dt}y_2 = B(1 - y_2) - Cg(x_2)y_2 \tag{4}$$

Gated On-Activation:

$$\frac{d}{dt}x_3 = -A_3x_3 + Dg(x_1)y_1 \tag{5}$$

Gated Off-Activation:

$$\frac{d}{dt}x_4 = -A_4x_4 + Dg(x_2)y_2 \tag{6}$$

Normalized Opponent On-Activation:

$$\frac{d}{dt}x_5 = -A_5x_5 + (E - x_5)x_3 - (x_5 + F)x_4 \tag{7}$$

Normalized Opponent Off-Activation:

$$\frac{d}{dt}x_6 = -A_6x_6 + (E - x_6)x_4 - (x_6 + F)x_3 \tag{8}$$

Total On-Activation:

$$\frac{d}{dt}x_7 = -A_7x_7 + G[x_5]^+ + L\sum_{k=1}^{n} S_k z_{k7} \tag{11}$$

Total Off-Activation:

$$\frac{d}{dt}x_8 = -A_8x_8 + G[x_6]^+ + L\sum_{k=1}^{n} S_k z_{k8} \tag{12}$$

On-Conditioned Reinforcer Association:

$$\frac{d}{dt}z_{k7} = S_k[-Hz_{k7} + K[x_5]^+] \tag{13}$$

Off-Conditioned Reinforcer Association:

$$\frac{d}{dt}z_{k8} = S_k[-Hz_{k8} + K[x_6]^+] \tag{14}$$

On-Output Signal:

$$O_1 = [x_5]^+ \tag{15}$$

Off-Output Signal:

$$O_2 = [x_6]^+, \tag{16}$$

pole circuit can describe the association between a CS with the onset and the offset of a reinforcer, a specialized gated dipole is needed to explain secondary inhibitory conditioning. Secondary inhibitory conditioning consists of two phases. In phase one, CS_1 becomes an excitatory conditioned reinforcer (e.g., source of conditioned fear) by being paired with a US (e.g., a shock). In phase two, the offset of CS_1 can generate an off-response which can condition a subsequent CS_2 to become an inhibitory conditioned reinforcer (e.g., source of conditioned relief). In order to explain secondary inhibitory conditioning, a gated dipole circuit must also contain internal feedback pathways, i.e., it should be recurrent. In addition, such a recurrent gated dipole must be joined to a mechanism of associative learning. The total circuit that we have analyzed is called a READ circuit, as a mnemonic for Recurrent Associative gated Dipole (Figure 9).

where the notation $[xi]+$ denotes a linear signal above the threshold value zero; that is, max $(xi,\ 0)$.

In the equations, I denotes the tonic arousal level, J the US input, Sk the kth CS, zk_7 and zk_8 the association of the kth CS with the on- and the off-response, respectively. A,B,C,D,E,F,G,H,K, and L are parameter values, which were kept constant for all simulations. When $E = F$, x_5 and x_6 compute an opponent process and a ratio scale at the same time. Thus one key property of the READ circuit is associative averaging, rather than summation.

6. Opponent Extinction by Dissociating Long Term Memory Read-In and Read-Out at Dendritic Spines

A second key property of the READ circuit has been called *opponent extinction.* Although passive memory decay does not occur in the parameter ranges which we used, when the net signals in the on- and off-channels are balanced, then $x_5 = 0 = x_6$, and therefore zk_7 and zk_8 approach 0. The LTM traces hereby continually readjust themselves to the net imbalance between the on- and off-channels. Opponent extinction avoids the possible saturation at maximal values of both LTM traces zk_7 and zk_8.

A third key property of the READ circuit is a dissociation between read-in and read-out of long-term memory (LTM), as in Figure 10. For example, in the on-channel, read-out is proportional to $[x_7]+$, whereas read-in is proportional to $[x_5]+$. Grossberg (1975) proposed that such dissociation can be physiologically implemented by assuming that synaptic plasticity occurs at the dendritic spines of neural cells. Signal $[x_5]+$ is assumed to cause a global potential change that invades all the spines inducing plastic changes throughout the dendritic column, as in equation (13). However, due to the geometry and electrical properties of the dendritic tree, an input that activates a particular dendritic branch may not be influenced by inputs that activate different dendritic branches. Activation at a particular dendritic branch would produce local potentials that propagate to the cell body where they influence axonal firing via potential x_7 in equation (11).

7. Computer Simulations of Primary and Secondary Conditioning

This section summarizes computer simulations in different classical conditioning paradigms. Although the simulations show the competence of the READ circuit in these paradigms, additional neural machinery (such as the ART circuit in Figure 1) is necessary to explain some difficult conditioning data.

Excitatory primary conditioning. Because the CS is presented in the presence of the US, it becomes associated with the on-response. Variable CS_1-ON describes conditioning of the LTM trace z_{17} within the pathway from the sensory representation of CS_1 to the onchannel. After 10 acquisition trials, presentations of CS_1 alone do not cause extinction of the CS_1-ON association (Figure 11). As explained later in the text, forgetting of CS_1-ON associations is due to the acquisition of CS_1-OFF associations.

Inhibitory primary conditioning. Because the CS is presented after the US offset, it becomes associated with the off-response. Variable CS_1-OFF describes conditioning of

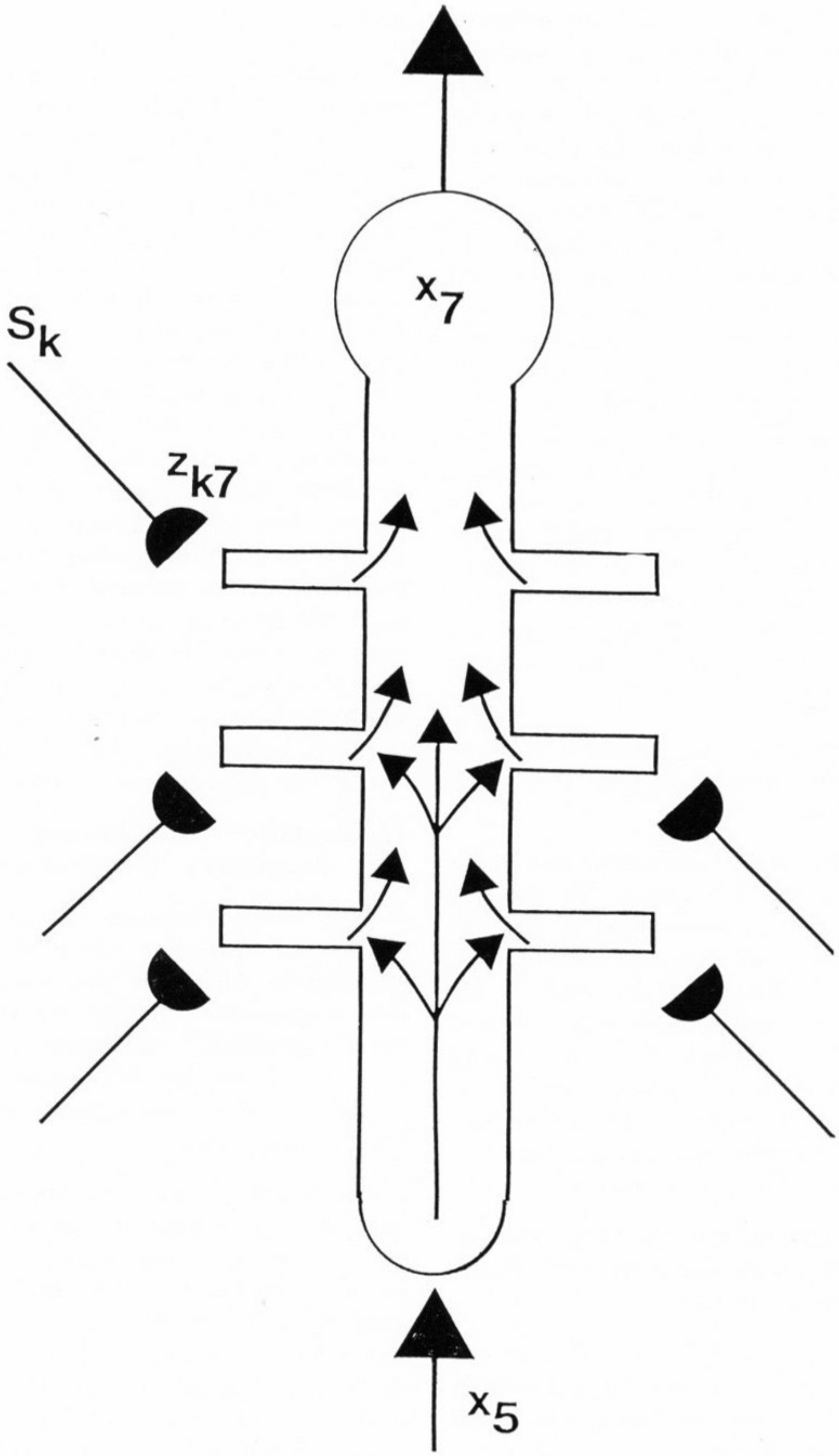


Fig. 10. — *A possible microarchitecture for dissociation of LTM read-in and read-out: Individual LTM-gated sensory signals* $S_k z_{k7}$ *are read-out into local potentials which are summed by the total cell body potential* x_7 *without significantly influencing each other's learned read-in. In conrast, the input signal* x_5 *triggers a massive global cell activation which drives learned read-in at all active LTM traces abutting the cell surface. Signal* x_5 *also activates the cell body potential* x_7.

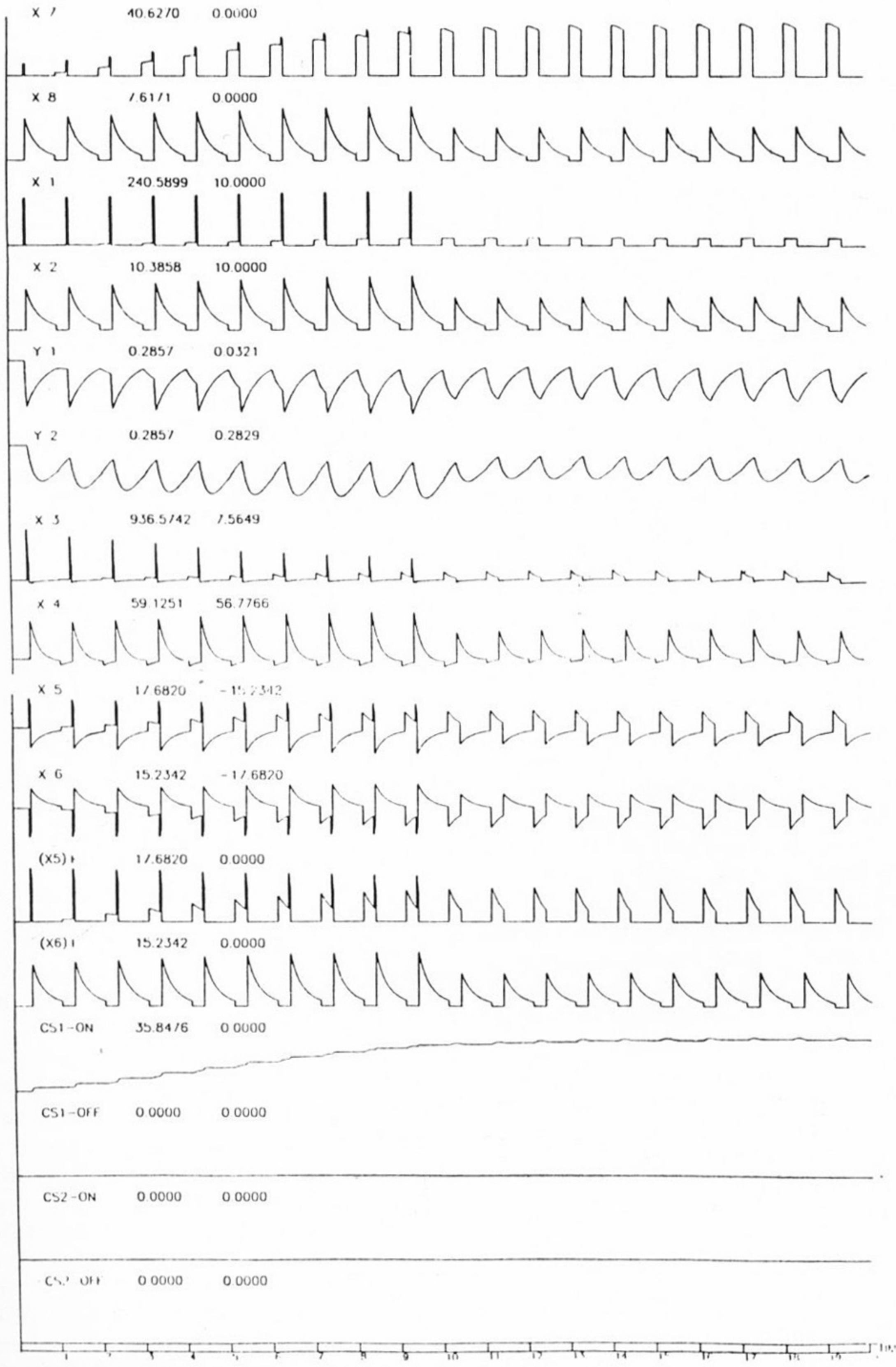


Fig. 11. — *Computer simulation of primary excitatory conditioning and extinction with slow habituation and large feedback in a READ I circuit:* CS_1 *is paired with the US during the first 10 simulated trials, and* CS_1 *is presented in the absence of the US in the next 10 simulated trials. The numbers above each plot are the maximum and minimum values of the plot. Parameters are* A = *1*,B = *.005*,C = *.00125*, D = *20*,E = *20*,F = *20*,G = *.5*,H = *.005*,K = *.025*,L = *20*,M = *.05*.

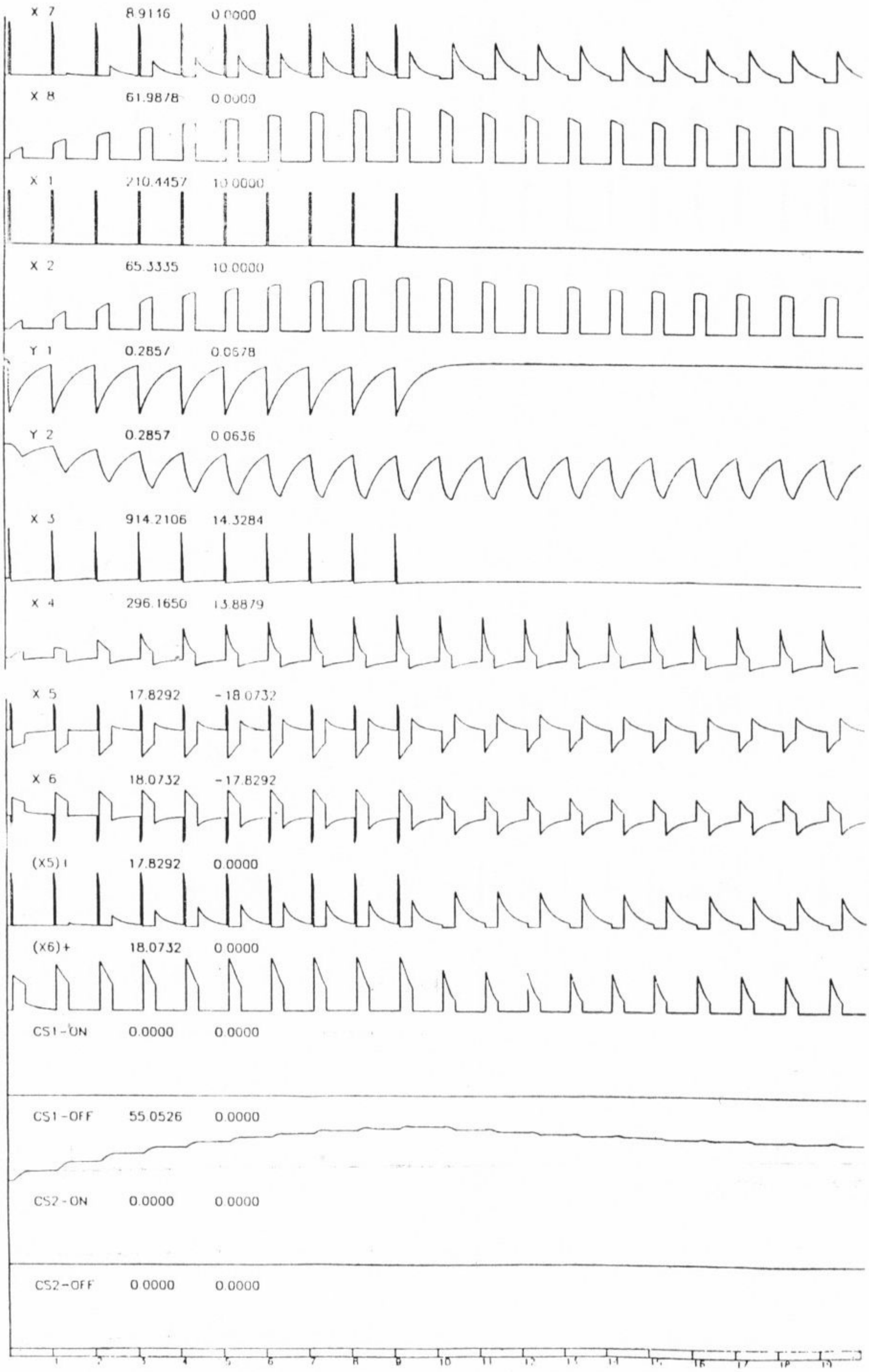


Fig. 12. — *Computer simulation of primary inhibitory conditioning and extinction with slow habituation and large feedback in a READ I circuit:* CS_1 *is presented after the* US *offset during the first 10 simulated trials, and* CS_1 *is presented in the absence of he* US *in the next 10 simulated trials. The same parameters were used as in Figure 11.*

the LTM trace z_{18} within the pathway from the sensory representation of CS_1 to the off-channel After 10 acquisition trials, presentations of CS_1 alone cause the CS_1-OFF association to relax to a persistent remembered value (Figure 12). As explained later in the text, forgetting of the CS_1-OFF association is due to the acquisition of CS_1 -ON associations.

In Grossberg and Schmajuk (1987), the following types of secondary conditioning phenomena are also simulated:

Excitatory secondary conditioning. The LTM trace CS_1-ON grows during the first 10 trials and is then used to induce the growth of the LTM trace CS_2-ON during the next 10 trials.

Inhibitory secondary conditioning. The LTM trace CS_1-ON grows during the first 10 trials and is then used, by presenting a CS_2 after CS_1 offset, to induce the growth of the LTM trace CS_2-OFF during the next 10 trials.

8. Qualitative Explanations of Extinction and Non-Extinction Data

This section presents qualitative explanations for some difficult conditioning data that require additional neural machinery, such as STM attentional modulation and STM reset by expectancy mismatch by an ART circuit.

Excitatory conditioning and extinction When a CS is paired with an aversive US on successive conditioning trials, the sensory representation S_1 of CS_1 is conditioned to the drive representation *Don* corresponding to the fear reaction, both through its conditioned reinforcer path $S_1 \longrightarrow Don$ and through its incentive motivational path $Don \longrightarrow S_1$. As a result, later presentations of CS_1 tend to generate an amplified STM activation of S_1, and thus CS_1 is preferentially attended. Due to the limited capacity of STM less salient cues tend to be attentionally blocked when CS_1 is presented.

As the cognitive-motivational feedback loop $S_1 \longrightarrow Don \longrightarrow S_1$ is strengthened during conditioning trials, S_1 is also associated to a sensory expectation of the shock within an ART circuit. During extinction. S_1 is presented on unshocked trials. Parameters of the READ cicuit are chosen to prevent passive decay of LTM traces from occurring on these trials. However, when the expected shock does not occur, a mismatch occurs with the learned expectation read-out by S_1, the STM activity of S_1 is reduced by the consequent STM reset, and an antagonistic rebound occurs in the off-channel of the READ circuit. Consequently, S_1 is associated to an antagonistic rebound at *Doff*. Because S_1 is smaller after reset than before, $S_1 \longrightarrow Doff$ associations take place at a slower rate than during conditioning. After several learning trials, however, the pathway $S_1 \longrightarrow Doff$ is as strong as the $S_1 \longrightarrow Don$ pathway, and opponent extinction occurs.

Inhibitory conditioning and non-extinction. Suppose that CS_1 has become a conditioned excitor, and that CS_1 and CS_2 are presented together in absence of the US. When CS_1 and CS_2 are simultaneously presented (Figure 13), S_1's activity is amplified by positive feedback through the strong conditioned $S_1 \longrightarrow Don \longrightarrow S_1$ pathway. As a result of the limited capacity of STM, the STM activity of S_2 is blocked at time T_1. When the expected US does not occur at time T_2, the mismatch with S_1's sensory expectation causes both S_1 and S_2 to be reset, and S_1's STM activity decreases while S_2's STM activity increases. Due to S_1's decrease, a rebound occurs at *Doff*. Consequently, the unexpected nonoccurrence of the shock enables S_2 to become associated with *Doff* in both the pathways $S_2 \longrightarrow Doff$ and $Doff \longrightarrow S_2$. These are the primary cognitive-motivational conditioning events that turn CS_2 into a conditioned inhibitor.

According to the READ circuit, when presented alone the conditioned value of $CS_2 \longrightarrow Doff$ persists. No further extinction occurs because the CS_2 sensory expectation predicts the absence of the US. Thus when presented alone, CS_2 does not disconfirm its sensory expectation, and S_2's STM activity is not reset.

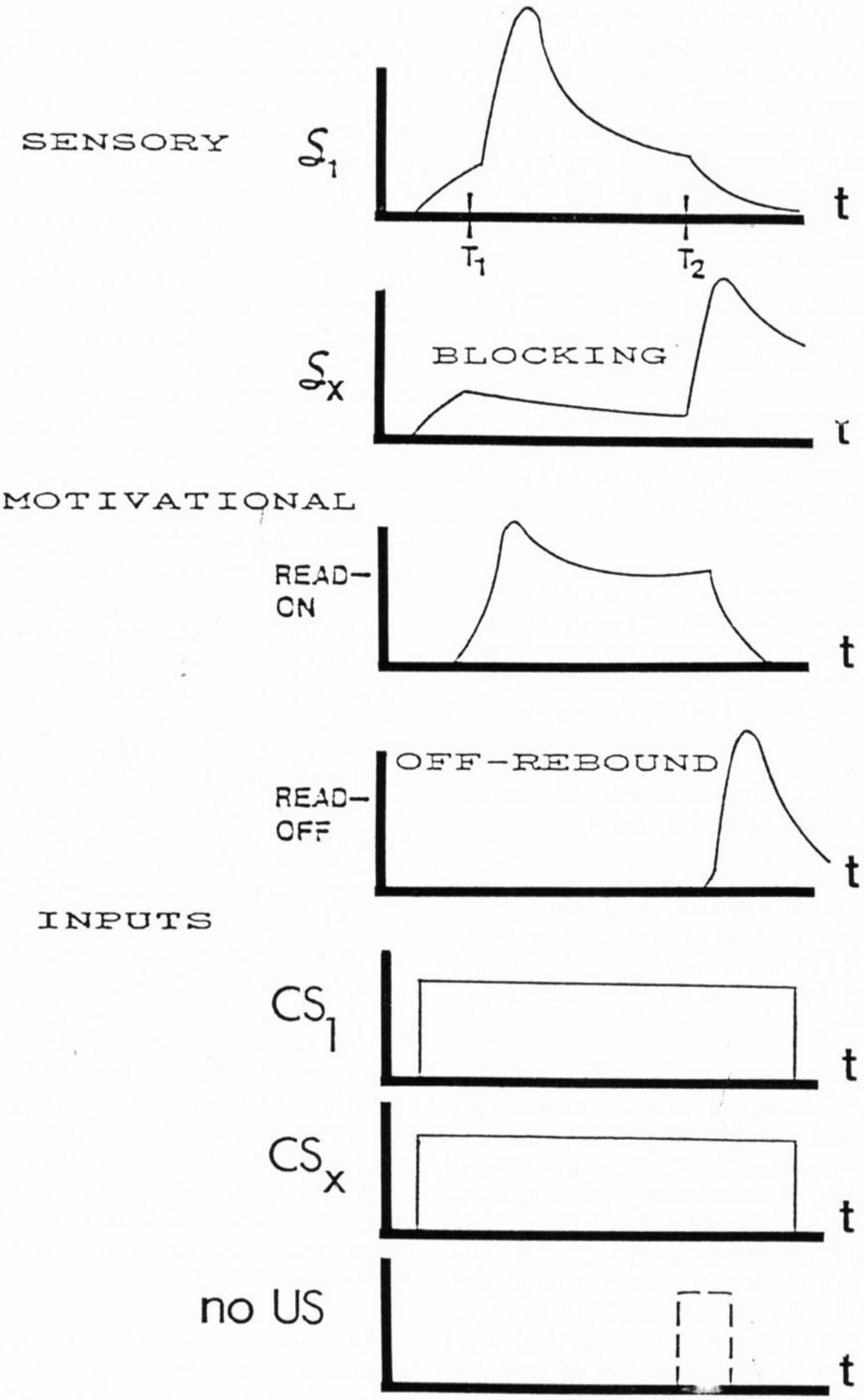


Fig. 13. — *Presentation of* CS_1 *and* CS_2 *when* CS_1 *has become a conditioned excitor and the compound stimulus is followed by no-shock: During the no-shock interval between times* T_1 *and* T_2 S_1 *is actively amplified by positive feedback and* S_2 *is blocked. Nonoccurrence of the expected shock causes both* S_1 *and* S_2 *to be reset.* S_1*'s STM activity decreases and* S_2*'s STM activity increases. Due to* S_1*'s increase,* Don *also decreases, hereby causing a rebound at* Doff. *This rebound becomes associated with the increased activity of* S_2.

SUMMARY

At least four types of learning processes are relevant in the present paper: learning of conditioned reinforcement, incentive motivation, sensory expectancy, and motor command. These several types of learning processes, which operate on a slow time scale, regulate and are regulated by rapidly fluctuating limited capacity STM representations of sensory events. The theory suggest how nonlinear feedback interactions among these fast information processing mechanisms and slow learning mechanisms participate in different conditioning paradigms, and actively regulate learning and memory to generate predictive internal representations of external environmental contingencies.

RESUMEN

Por lo menos cuatro tipos de procesos de aprendizaje tienen relevancia en el presente escrito; aprendizaje con refuerzo condicionado, motivación incentivada, expectativa sensorial e imperativo motriz. Estos diferentes tipos de procesos de aprendizaje que actaún con ritmos de lentitud en el tiempo, regular y son regulados por rápidos fluctuantes de capacidad limitada, memoria de corto plazo, representaciones de eventos sensoriales. La teoría sugiere como interacciones retroactivas no alineadas entre estos mecanismos procesantes de rápida información y mecanismos de lento aprendizaje que participan en diferentes modelos condicionados y activamente regular aprendizaje y memoria para generar representaciones internas predictivas de contingencias ambientales externas.

RÉSUMÉ

Il est présent catre types de processus d'apprentissage importants, dans cette étude; aprentissage avec renfort conditioné; mobivantion incentivée; attente sensorielle; motrietétimperative.

Ces diferents types de procedés agisent avec des rytmes de durées variables et sont regets pardes fluctuations de capacité lemitée, mémoire a court terme, representations de faits sensoriels.

La theorie sugiere comme interactions rétroactives mon alignées avec ces processus d'information rapide at mécanismes d'apprentissage lent, qui interviement dans différents modéles conditionés et activement régulés four génére des representations internes prédictives a partir de contingences ambientales externes.

ZUSAMMENFASSUNG

Mindestens vier Typen von Lernprozessen sind von Bedeutung in dieser Schrift: Lernen mit bedingter Nachhilfe, aus anreizender Motivierung, sinnlicher Erwartung und motorischem Antrieb. Diese unterschiedlichen Typen des Lernprozesses, die sich in zeitlich langsamen Mass abwickeln. geordnet und sind geregelt durch rasche Schwankungen begrenzter Fähigkeit, Kurzzeitgedächtnis und Vorstellungen sinnlicher Ereignisse. Die Theorie legt nahe, dass nicht geradlinig rückkoppelnde Zwischenwirkungen an diesen Schnellerkenntnis liefernden Mechanismen und Mechanismen des langsamen Lernens teilhaben an verschieden bedingten Beispielen und aktiv Lernen und Gedächtnis regeln um lehrhafte innere Vorstellungen zu erzeugen von Zufälligkeiten der äusseren Umgebung.

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Can Memories be Encoded Across Generations?

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Theories of memory should encompass not only individual acquisition through life-times but also encoding through generations. Certain behavioral uniformities are evident through generations of the human species. Consider, for example, the basic emotional displays and epileptic propagation patterns. Are these memories? The definition is difficult. The term, memory, can probably not be restricted to the conscious awareness of prior events but should include impresses on the nervous system acquired and automatized below awareness; impresses arising from pathways of facilitation genetically based.

Studies on familial psychopathology indicate that children and their parents, at times their grandparents, may display ideas or actions which are identical. Endowed with strong emotion and not easily dispelled from consciousness, these may be called "imperative" ideas and actions (Epstein 1972). They do not seem to be imitative or copied. These highly charged ideational complexes may be viewed as memories which are somehow encoded to appear in the next generation.

CASE EXAMPLES

Case examples of imperative ideas and actions gained from the literature are now cited.

Ideas and actions in the sexual deviations are insistent, highly emotionally charged and usually associated with genital arousal.

Liakos (1967) reported fetishism-transvestism in a 61 year male who always had the tendency to wear female clothes and, at age 59, started to collect his wife's clothing. underwear and dresses. His son, age 17, was repeatedly found in bed wearing female underclothing and stated he "had the desire to dress himself in female clothes ever since he could remember". His other son was found at about age 27 to have a box of women's underclothes in his possession. At that age, he was also charged with stealing woman's brassieres and stockings from a clothing line.

Buhrich (1977) described a man whose first transvestite (cross-dressing) experience occurred at age 4. At age 16, he began to cross-dress fully. The cross-dressing was associated with sexual arousal and masturbation. He had fantasies of being a woman. The man's son was "attracted to female clothes for as long as he could remember". Until puberty, the son cross-dressed two to three times a week. During adolesence, while cross-dressed, he usually masturbated. He also had fantasies of being a woman. The man was "astonished" when he recently learned of his father's transvestism.

Krueger (1978) described a man who first cross-dressed at age 16. He would don women's underwear. Erections occurred while cross-dressing. At age 16 or 17, he "began collecting women's clothes, primarily underpants and bras...". A son began cross-dressing at age 13 with erections. The cross-dressing "involved women's underpants, bras and pantyhose". A second son began cross-dressing at about age 14. Erections and masturbation occurred while cross-dressing. A third son, who had epilepsy, type unstated, "thought about cross-dressing from time to time" and "masturbated and ejaculated into the women's clothes". The fourth son was arrested at age 11 for "trying on women's clothes at a garbage dump". He was arrested again at age 14 "when he wore women's clothes on the street". Erections occurred when he crossed-dressed.

Comings and Comings (1982) reported exhibitionism in a man with accompanying Tourette's syndrome. The exhibitionistic urges, beginning at age 15, were dominating and led to two arrests. One night, he saw his 9 year old son exposing himself.

Shapiro (1980) described a man with an episode of exhibitionism in adolescence. His 16 year old son was hospitalized for episodes of exhibitionism.

The suicidal thought and act, indeed any imperative idea of death, is emotionally laden and, therefore, insistent. Zaw (1981) described male identical twins: the first committed suicide by shooting at age 21, the second committed suicide by shooting at about age 30. The twins' father cut his wrists and throat at about age 53 but survived. The twins' mother had a depressive illness but did not attempt suicide. The twins' paternal and maternal grandfathers both committed suicide by cutting their throats.

The maternal grandmother of the proband described by Dilsaver and White (1986) displayed marked self-reproach and "wanted to die". The mother of the proband "had an overwhelming fear that she might die". The proband, a 17 year female, had "recurrent thoughts of death and suicide". A brother of the proband had a "preocupation with death and suicide"; another brother had depressive episodes.

Phobias are heavily fear-laden, and with the appropriate external stimulus, may dominate the psyche. Rapp and Thomas (1982) reported a 47 year woman who had agoraphobia since age 22. Her daughter, 23 years of age, for the past six months "had been unable to go anywhere alone without experiencing significant anxiety".

In Tourette's syndrome, the complex stereotyped action (tic) and affectively-laden vocalization or idea (coprolalia or other obsessional thought) are insistent, defying voluntary control. Lucas (1973) reported a woman whose skipping movements and involuntary vocalisations began at age 8. Her son developed skipping mannerisms at age $4\frac{1}{2}$ and vocalizations a few months thereafter.

Sanders (1973) reported a man who struggled against coprolalia. His son, age 23, also had an "intrusion of obscene words into consciousness" which required suppression.

Friel (1973) reported both a mother and son with tics, including vocal grunts. Eisenberg et al. (1959) reported both a father and son with facial grimicing and eye-blinking. Dunlap (1960) also reported a father and son with eye-blinking tic.

DISCUSSION

The identity of transgenerational ideation and action is supported by more general studies of familial psychopathology. Gaffney, Lurie and Berlin (1984) found sexual deviancy in 18.5 per cent of the families of paraphiliacs, but the deviancy among the families of pedophiles consisted of pedophilia. Lewis (1935) studied the parents of 50 obsessional patients; of the 100 parents, 37 showed "pronounced obsessional traits". Roy (1983) found that among 243 patients with a family history of suicide, 48.6 per cent had attempted suicide. Solyom et al. (1974) found that of 47 phobic patients, 30.9 per cent had a phobic mother and 6.3 per cent a phobic father. Shapiro et al (1978) found that in 139 patients with Tourette's syndrome, 34.5

per cent had a history of tics in family members. Similar dream types across generations have also been described (Epstein and Collie 1976).

In the majority of the case examples cited above, it is unlikely that imitation (identification) is a factor. Another mode of transmission of the ideational and action complexes is more likely and here one invokes a biologic, a genetic explanation. Should this be the case, it is not clear whether a general disposition, e.g. an affective tendency, is inherited or whether a more specific ideational or action complex is transmitted. Could the genetic mechanism, in some instances, follow a Lamarckian mode, i.e. the preceding generation acquires an affectively laden ideational or action complex which is encoded in an unique fashion permitting its passage to the next generation? The phrase, "unique fashion", implies that strongly affectively laden complexes are encoded in a particular physicochemical manner which endows them with a preferential (hierarchically superior) impress. This concept of unique coding is supported by the occasional epileptogenic capacit of memories of extremely traumatic events (Epstein 1964). Whether the memory of an extremely traumatic event can be transmitted across generations is highly speculative (Epstein 1982) but is not out of keeping with the concepts of certain early memory theorists such as Semon (1921). Consideration of this question has important implications for the nature of the memory trace.

SUMMARY

Studies of familial psychopathology, specifically of "imperative ideas and actions", indicate a frequent transgenerational occurrence. These affectively highly charged complexes (memories) are encoded, it is speculated, across generations by a genetic mechanism.

RESUMEN

Estudios de psicopatología familiar, expecíficamente de "ideas y acciones imperativas", indican una ocurrencia frecuente transgeneracional. Estos complejos en alto grado afectivamente marcados (memorias) son codificados, se ha especulado através de generaciones por un mecanismo genético.

RÉSUMÉ

Les études des troubles psycho-pathologiques familiaux montrent que ceux-ci, en particulier les pulsions avec ou non passage à l'acte, possédent une fréquente incidence transgénérationnelle. L'hypothèse est proposée que ces complexes (mémoires) qui possédent une haute charge affective, sont codés au travers des générations par un mécanisme génétique.

ZUSAMMENFASSUNG

Untersuchungen ueber familiaere Psychopathologie besonders ueber "Imperative Ideen und Aktionen" ergeben ein haeufiges transgererationelles Auftreten. Diese hochgradig affektiv markierten Komplexe (Gedaechtnisdaten) sind kodifiziert, wie man glaubt, durch einen ueber Generationen wirksamen genetischen Mechanismus.

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MSH/ACTH 4-10 and Aging Effects on Delayed Response Performance in Rats

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INTRODUCTION

Delayed response performance has been used as a measure of short term memory of visual stimuli in animals and humans beginning in 1913 (Hunter, 1913). Honzik developed an opaque door modification which eliminated the confound of postural positioning allowing for more accurate assessment of delayed response performance in animals (Honzik, 1918). From that time until quite recently the delayed response design and various analogs of the paradigm have been used to determine how long various species can successfully recall the correct properties (location, color, size, etc.) of previously presented stimuli in a normal, undrugged state (Cowles, 1940). Relatively few explicit theoretical models based upon learning and performance exist for the delayed response paradigm. However, Cowles' (1940; 1941) "early discrimination" model and Fletcher's (1965) more recent "intra-trial, orienting response analysis" attempt to describe delayed response performance in terms of conventional learning theory. More important than the theoretical framework for the paradigm itself is the fact that the Hunter apparatus allows reliable assessment of a subject's delayed response performance (DRP). Up until quite recently, DRP has been neglected for use in assessing cognitive effects of CNS acting drugs. Recently, Bartus and co-workers have used an indirect delayed response-matching to sample procedure in young and aged monkeys with great success in determining DRP (Bartus, 1978; Bartus, et al., 1982). Sub-primate species such as rodents may also prove to be useful in the investigation of psychopharmacological agents, aging and memory in the delayed response design.

The discovery that MSH/ACTH peptides delay conditioned avoidance response extinction began a vast amount of behavioral research regarding these peptides (de Wied, 1965; Miller and Ogawa, 1962). The more recent findings involving the identification of endogenous opiate receptor ligands (endorphins) and that some of these peptides were synthesized from the same precursor molecule as the MSH/ACTH peptides added greatly to the already huge amount of data regarding opiomelanocortinergic peptides.

While discussing the behavioral actions of ACTH-type peptides it will be assumed that MSH and ACTH have similar, if not identical activities (Kastin, et al., 1973. The only difference noted was an instance of contrasting effects of MSH and an ACTH analog, one which considerable controversy surrounds (Dornbush, Shapiro and Freedman, 1981; de Wied, Witter and Greven, 1976), on locomotor activity in the rat (File, 1981). In addition to this problem in assessing MSH/ACTH research, the non-steroid generating MSH/ACTH peptide fragments have complicated the field (Beckwith and Sandman, 1978. This complication is probably for the best for several reasons. First, MSH/ACTH 4-10 would seem to have potent behavioral effects similar to ACTH or MSH (Beckwith and Sandman, 1982. Additionally, the lack of steroid generating effects eliminates one of the serious confounds when analyzing the components of the effect of MSH /ACTH peptides on behavior or other dependent variables.

A controversial point in behavioral neuropeptide research is the route of administration (Kastin, Coy, Schally and Miller, 1978). The penetrance of the blood-brain barrier by peripherally administered peptides is now widely accepted due to behavioral, physiological and biochemical evidence (Banks, Kastin and Coy, 1982; Kastin, et al., 1979; Kastin, et al., 1978; Miller, et al., 1981).

In asymptomatic elderly MSH/ACTH 4-10 has been demonstrated to have varying degrees of cognitive improvement (Ferris, Reisberg and Gershon, 1980; Miller, et al., 1980). This may be compared to studies investigating the effect of MSH/ACTH 4-10 on memory and attention in cognitively symptomatic elderly which have yielded mixed results (Abuzzhab, Will and Zimmermann, 1982; Branconnier, Cole and Gardos, 1979; Ferris, Reisberg and Gershon, 1980). These peptides have also been demonstrated to be of some social benefit in the retarded population (Sandman, Walker and Lawton, 1980). Such clinical studies should be continued with better controls and using more consistent measurements of cognition, memory or attention.

Animal studies have found that MSH/ ACTH peptides delay extinction of conditioned avoidance responses in rats (de Wied, 1965). Visual sensory information processing has also been found to be enhanced in models that use discrimination as a dependent factor (Beckwith, Sandman and Kastin, 1976; Sandman, et al., 1977). While MSH/ACTH fragments have been found to lack effects on avoidance acquisition, certain analogs of these peptides have demonstrated acquisition enhancement properties (Martinez, et al., 1979). Taking a different approach, some investigators have found MSH/ACTH peptides are able to partially reverse experimentally induced amnesia in rats (Mactutus, Smith and Riccio, 1980; Rigter, van Riezen and de Wied, 1974). Amnesia can be induced by various methods. Popular methods include hyperthermia, oxygen deprivation and electroconvulsant treatment. A few laboratories use appetitive learning as a paradigm for assessing the effects of MSH/ACTH peptides of memory and learning. In these designs the opiomelanotropic peptides enhance acquisition or improve performance in these tasks (Bartus, Dean and Beer, 1982; O'Reilly, Coleman and Kg, 1983). In the studies involving aged or cognitively impaired animals, Bartus and his group have demonstrated that MSH/ACTH fragments can improve memory and retention in some, but not all, animlas (Bartus, Dean and Beer, 1982).

In both animal and man MSH/ACTH has been shown to have effects on the electrical activity of the brain (Pfaff, Silva and Weiss, 1971; Sandman, et al., 1971). Contrasting studies are fewer but none the less present in human studies, demonstrating the controversy which is present in the cross-species generalization of peptide results (Sannita, Irwin and Fink, 1976). A similar situation exists in the area of MSH/ACTH effects on cognition in humans. While it is accepted without question that MSH/ACTH can retard the extinction of the conditioned avoidance response in animals, this cannot be stated in the human case (Miller, et al., 1976; 1977). Differential results regarding the effects of different peptide fragments on attentional processes have been obtained (Flood, et al., 1976; Sandman, Kastin and Miller, 1977). These differences in potency of the different peptide fragments do not seem to be pronounced to such a degree in animal studies as in found in human subject experiments (Dornbush and Nikolovski, 1976; Miller, et al., 1976; 1980). When taken as a whole, the bulk of the data reported does tend to support the attentional enhancement properties of MSH/ACTH 4-10 as operationally defined by standard psychometric tests such as the Sternberg and continuous performance task (CPT).

Rich and Thompson have demonstrated that rats trained to perform an avoidance response in reaction to a visual stimulus fail to demonstrate retention of this discriminative avoidance response after gross disruption of the septohippocampal neuronal system (Rich and Thompson, 1965). This finding has been repeatedly demonstrated in experimental situations (Moore, 1964; Peretz, 1960). Bilateral damage to either the septofornix area, hippocampus, anterior thalamus or posteriolateral hypothalamus created such a deficit while bilateral lesions of the anterior hypothalamus or rostral medial forebrain bundles did not. From this Rich and Thompson concluded a "priority of the thalamic route over the hypothalamic route in the functional activation of the hippocampal -septal system". It is interesting to note that lesions in the dorsal hippocampus abolishes the behavioral actions of MSH/ACTH peptides in the avoidance procedures (Wimersma Greidanus and De Wied, 1976). However, this location does not appear to be singly responsible for the actions of these neuropeptides as lesions of the parafascicular nuclei (Wimersma Greidanus, et al., 1974) and amygdaloid nuclei (Wimersma Greidanus et al., 1979) also appear to block some of the behavioral effects of these substances.

A recent study postulates that the septohippocampal neurons, which are cholinergic in nature, are regulated trans-synaptically by opiomelanocorticotropic peptides (Botticelli and Wurtman, 1982). It was found that beta-endorphin administered intraventricularly (1,3,10, or 30 ug) or intraseptally (1 ug), increased levels of ACh, while ACTH 1-24, injected similarly, decreased levels of the neurotransmitter in the dorsal hippocampus. While MSH/ACTH 4-10 was without measureable effect in this study, it is possible that the effect of this peptide on ACh neuronal functioning may transiently exist in a fully functioning, non-anesthetized animal. This negative finding in regard to the effect of MSH/ACTH 4-10 of ACh transmission may not extend to situations or periods of measurement other than those specified in the Botticelli and Wurtman study. In addition, technical problems involving the assay of ACh have plagued researchers for decades. Small, but significant increases in ACh release in brain could easily be obscured due to the large variance of measurement caused by the assay procedure.

Peripheral neuronal systems and hormone receptors are thought to predate the development of the central nervous system (Ross, et al., 1982). This theory has been used by researchers to investigate ACh pathologies of the CNS by observing correlated peripheral ACh measures (Nadi, Nurnberger and Gershon, 1984). These resent developments along with the numerous similarities of the neuromuscular junction to central ACh synapses suggest that peripheral ACh findings may be replicated in the CNS to varying degrees.

The enhancement of cholinergic transmission has been found to exist in the neuromuscular system (Strand and Smith, 1980). Strand and co-workers have demonstrated increased amplitude of muscle action potentials (Strand, et al., 1975), increased muscle contraction amplitudes and delays in muscle fatigue after administration of MSH/ACTH peptides (Strand, et al., 1975; Strand, et al., 1976). Their work and the result of others in the field suggest that these effects are modulated by increases in ACh release by the motor neuron, not by local effects at the motor endplate (Johnston, et al., 1983). The initial site of the resulting enhanced ACh release is most likely the cell body which originates in the spinal column. Thus, MSH/ACTH peptides have been demonstrated to alter cholinergic release and functioning in the CNS and periphery via a cell (motorneuron) whose body lies in the CNS. As yet no statement regarding this observation and its implications in cognitive processes has been made.

Using a classical, two choice version of the Hunter delayed response apparatus implementing the Honzig opaque door modification, the effects of age and several psychotropic agents on DRP was investigated.

While Hunter initially reported a three to five second retention span in rodents, a maximum retention of 8 sec was recorded by Honzik after introducing the opaque door modification to the Hunter paradigm. Hunter experienced postural cueing in some animals, a phenomena which was removed almost totally by the placement of an opaque door between the subject and the goal areas. Thus, the opaque door modification not only removed the postural cueing confound, but increased delay lengths. Honzik theorized that the removal of competing response stimuli by the opaque door was the cause of this increase (Honzik, 1931).

The effects of peripherally administered MSH/ACTH 4-10, alone and in conjunction with monoaminergic and cholinergic agents in young animals on DRP was assessed. Secondly, the effects of the natural aging process in rodents on DRP was measured. As the experimental animals approach 30 months of age, a second MSH/ACTH 4-10 - DRP dose response will be performed. Initially, it is hoped that the work of Bartus (1978; 1980) which used monkeys would be replicated with rodent subjects. In addition, the longitudinal study of DRP in animals explored the similarities of muscarine blockade to natural age-induced changes in DRP. By manipulation of the cholinergic system and measuring DRP over long segments of the animal's life-span this study constitutes an investigation of the cholinergic hypothesis of age-related memory alterations in a behavioral setting. The assessment of MSH/ACTH 4-10 on DRP may provide behavioral evidence supporting an adrenocorticotropin - ACh interaction in the CNS, as is found in the periphery. This interaction, if supported, may have functional implications in the cognitive effects of aging.

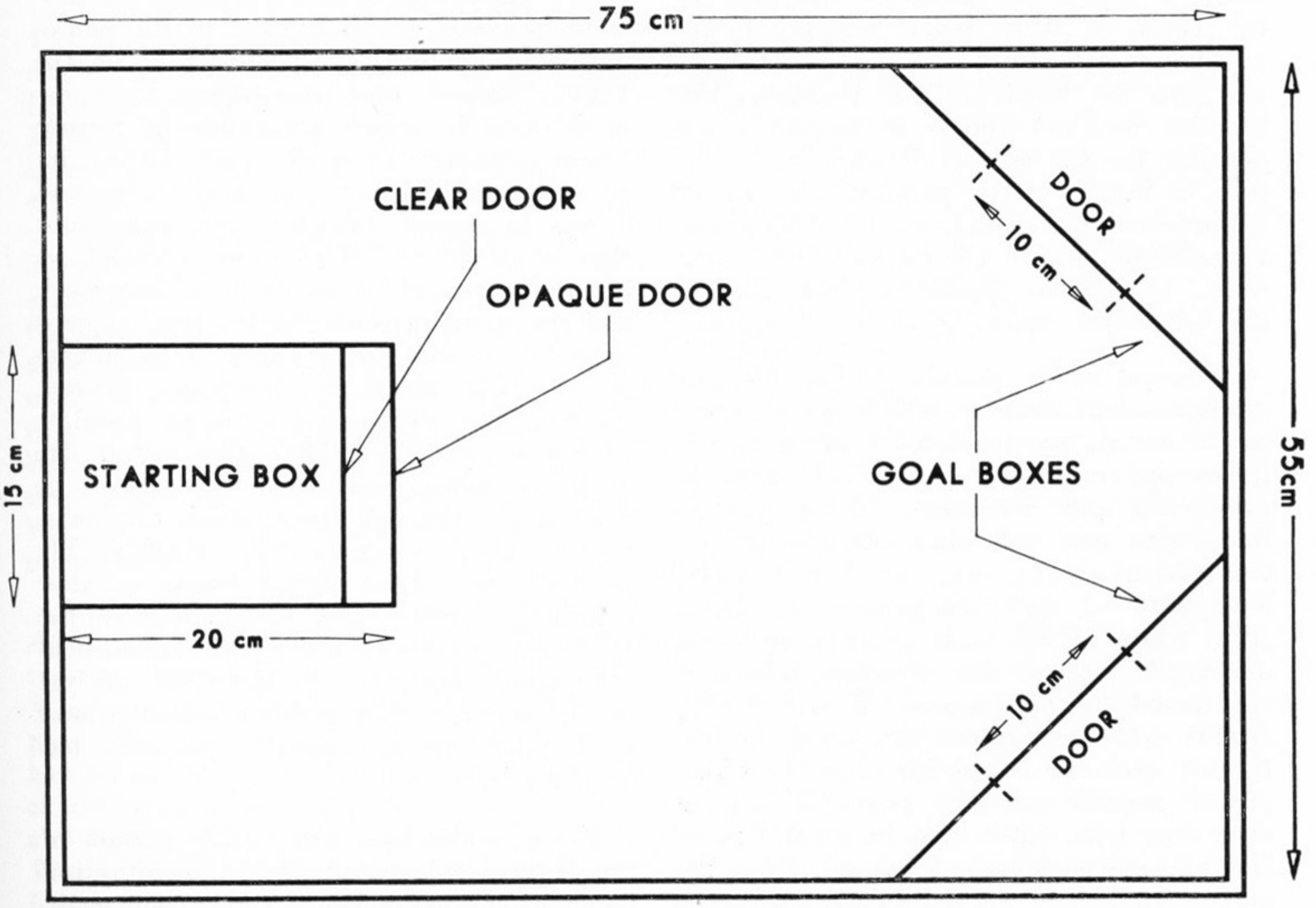


Fig. 1. — *Schematic of the hunter delayed reaction apparatus incorporating the honzik opaque door modification.*

METHOD

Six male Long-Evans, 70 day old rats (at the start of the study) were housed in stainless steel cages on a 12 hr light-dark cycle. Animals were maintained on Lab Chow at 85 % (275-350 g) of their ad libitum weight.

Delayed response performance (DRP) was assessed in a two choice Hunter apparatus (Hunter, 1913) with the Honzik opaque door modification (Honzik, 1931). The apparatus consisted of a 75 x 55 cm enclosure with a 20 x 15 x 15 cm starting box at one end and two goal boxes, located at the corners of the apparatus, at the other. The starting box was equipped with two vertically sliding doors, one of clear plexiglass on the inner side of the box and the other opaque, capable of preventing viewing of two goal boxes by the animal. To enter a goal box animals were first required to open a 10 x 10 cm door placed directly in front of each goal box. A light, capable of being illuminated by a 24 volt DC power source, was placed 2 cm over the center of each door.

Animals were shaped to exit the starting box and open goal doors to receive reward which was placed in goal boxes (approximately 0.2 g chocolate). Both goal boxes were supplied with reward to eliminate olfactory cues in door selection. Training consisted of allowing the animal to view the two goal doors, one of which had the light over it illuminated, through the clear plexiglass door from the starting box for 3 seconds. The opaque door was then lowered for a variable delay period while the illumination of the light was terminated, and both doors were then raised allowing the animal to choose one of the goal boxes. A correct response was made when the animal selected the goal box which had had the light illuminated above its door prior to the delay period. The incorrect door was bolted, preventing entry. Random alternation of correct goal boxes on a trial by trial basis was performed throughout the study.

Initial learning of the task was defined as four or more correct responses out of five trials at the 0, 2 and 4 sec delay periods for 16 out of 20 training days. A correct choice was defined as approaching and opening the correct goal door, and an incorrect choice consisted of approaching within 3 cm of the locked goal door or failing to make a choice within 3 seconds. Subsequent training was given with trials at 6, 8, 10 and 12 sec delays as well as the 0, 2 and 4 sec delay periods. A 15 sec pause existed between each trial. Testing consisted of five trials per delay period (0, 2, 4, 6, 8, 10 and 12 sec) per animal.

In a repeated measure design, animals received vehicle (0.01 mM acetic acid), MSH/ACTH 4-10 (47, 95, 190 and 285 ug/kg), atropine sulfate 2.0 mg/kg, strychnine sulfate 1.0 mg/kg, psysostigmine sulfate 0.5 mg/kg, methylpenidate HC1 2.0 mg/kg, and imipramine HC1 10.0 mg /kg. In addition, the following combinations were administered to the animals before DRP assessment: MSH/ACTH 95 ug/kg plus atropine sulfate 2.0 mg/kg. physostigmine sulfate 0.5 mg/kg plus atropine sulfate 2.0 mg/kg, methylphenidate HC1 2.0 mg/kg plus atropine sulfate 2.0 mg/kg, strychnine sulfate 1.0 mg/kg plus atropine sulfate 2.0 mg/kg, imipramine HC1 10.0 mg/kg plus MSH/ACTH 4-10 95 ug/kg and imipramine HC1 plus physostigmine sulfate 0.5 mg/kg. Doses listed were chosen after review of existing literature which demonstrated past success in producing reliable results with the substances to be tested (Bartus, 1978; Beckwith and Sandman, 1982; Brennan and Gordon, 1977; Lippa, et al., 1980). Pilot studies with at least half of the experimental population (n = 3) were performed with various doses of the agents tested in order to assess suitability in the DRP paradigm. Solutions were given intraperitoneally (IP) 1 hr before DRP testing. Between pharmacological treatments, animals were required to perform four days out of five, not significantly differing from baseline (vehicle) at any delay period. At least seven days passed between treatments regardless of animal performance.

After initial pharmacological treatments, animals were tested in the Hunter delayed response apparatus at least once per month for a period extending through 30 months of age. As in previous DRP testing situations, animals were required to maintain a consistent level of performance for 4 out of 5 pre-test training days. However, significant performance changes as compared to initial control DRP were accepted due to anticipated age-related alterations in performance. After completion of the longitudinal DRP assessment section of the study, animals were again given the various doses of MSH/ACTH 4-10 (vehicle, 47, 95, 190 and 285 ug/kg) before assessment of DRP. Again, animals were expected to perform in the delayed response apparatus in a statistically consistent fashion 4 out of 5 pre-DRP assessment days. In addition, free feeding weights were assessed every 4 months after the initial 10 month control. This assessment consisted of allowing animals free access to lab chow for 1 week prior to weighing. Proper assessment of free feeding weight allowed the accurate determination of 85 % ad libitum weight for the purpose of partial food deprivation. All testing in the delayed response apparatus was performed approximately 6 hr into the dark component of the light-dark cycle.

Data analysis for delayed response observations was performed using analyses of variance (ANOVA) with repeated measures. Separate analyses were performed for the initial pharmacological section, age-related changes in baseline data and the young vs. aged MSH/ACTH 4-10 dose-response section of the study. Drug related DRP and the effect of age on DRP were analyzed by two-way ANOVAs. The analysis for the MSH/ACTH 4-10 dose responses in animals when young and aged was accomplished with a three-way ANOVA with repeated measure. Data analysis for chances in ad libitum weight was performed via a one-way ANOVA with repeated measure. All post-hoc comparisons were performed using the Newman-Keuls method for treatment and/or cell mean comparisons. In addition, subject age was correlated with DRP starting at an age of 3 months and ending at 30 months of age. A planned comparison between DRP in 3 month controls, 30 month controls and animals after atropine sulfate administration was performed using a two-way analysis of variance with repeated measures. Correlations were performed using individual DRP scores and mean scores at each age interval. Trend analyses for MSH/ACTH 4-10 dose responses, young and aged, were performed using the respective age controls as baselines. Using the binomial distribution, greater than chance goal selection at a given delay period occurs above the 63.3 percentile (19 or more correct choices out of 30 trials) when a treatment ($n = 6$) is taken as a whole.

RESULTS

Although the final analysis for the MSH/ACTH 4-10 dose response included data from animals when young and old, the optimal peptide dose for the drug study section was selected after a dose response was obtained from animals when young. From this data it is clear that MSH/ACTH 4-10 at a dose of 95 ug/kg produced the most marked positive effect on DRP. Preliminary analysis of the MSH/ACTH 4-10 dose response yielded a significant difference between treatment conditions, $F(4,20) = 8.52$; $p = 0.0001$. Treatment/delay interaction was found not to be significant, $F(24,120) = 1.29$; $p = 0.1886$. Post-hoc analysis using the Newman-Keuls method revealed that MSH/ACTH 4-10 95 ug/kg did, in fact, differ significantly from control ($p < 0.01$). No difference was noted at a dose of 47 ug/kg and the two highest doses resulted in significantly impaired DRP ($p < 0.05$). Thus, MSH/ACTH 4-10 at doses of 195 and 285 ug/kg resulted in significantly worse delayed response performance in the Hunter paradigm when compared to control. While this inverse "U" dose response was not fully expected, it was clear that an optimal dose of MSH/ACTH 4-10 would have to be used if reversal of anticholinergic impairment of DRP was to be attempted. Using this preliminary data it was decided that MSH/ACTH 4-10 at a dose of 95 ug/kg would be used for combination with various agents in the drug study section.

Analysis of variance for the drug related data yielded a significant difference between treatments, $F(12,60) = 2.43$;

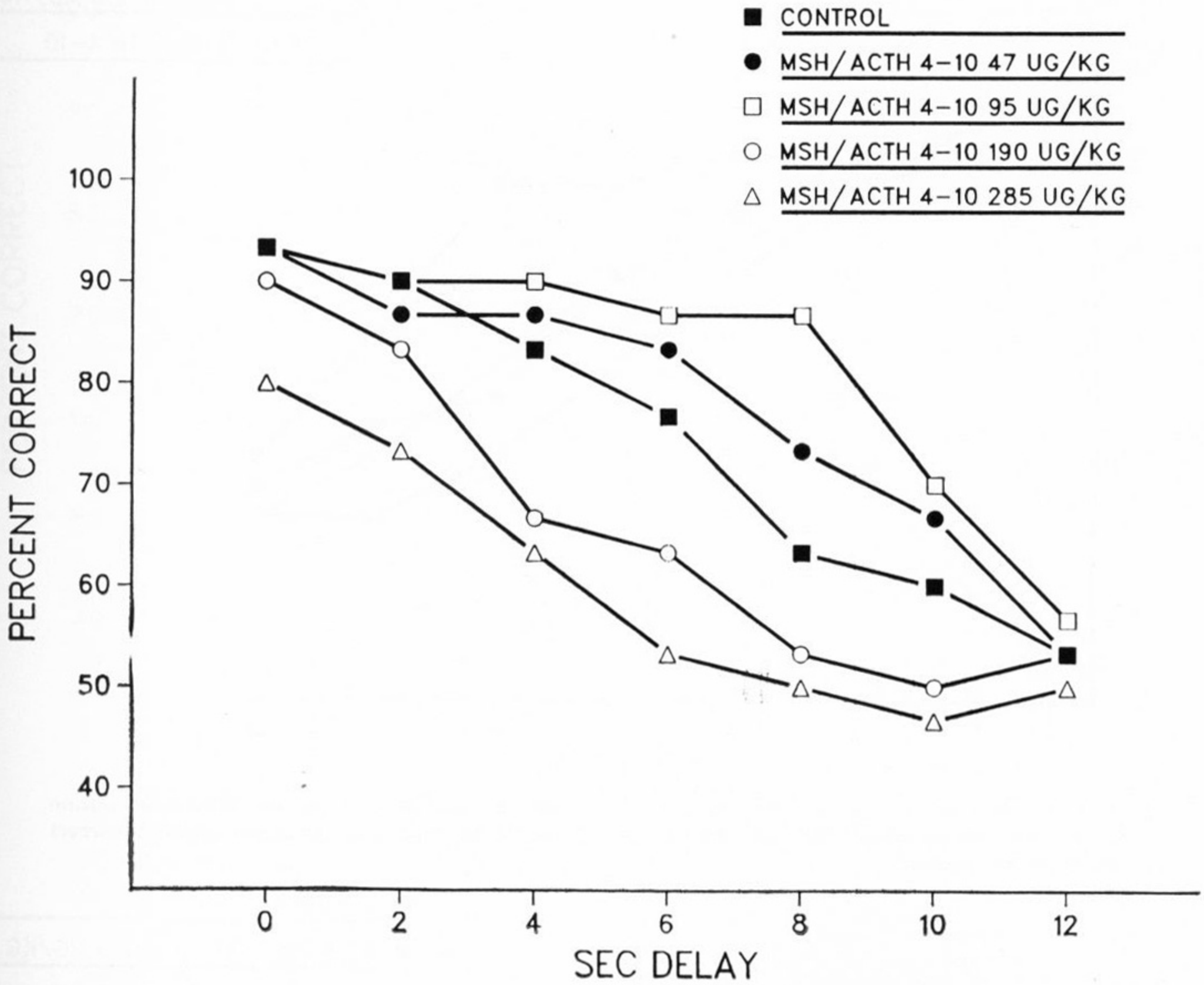


Fig. 2. — *Dose effect of MSH/ACTH 4-10 on delayed response performance in Young animals. Data expressed as mean percentage correct at each period of delay.*

$p = 0.0120$. The interaction between treatment and delay period was found to be non-significant, $F(72,360) = 1.13$, $p = 0.2391$. Post-hoc treatment comparisons indicated that atropine sulfate and imipramine HC1, at the doses given, significantly impaired DRP when compared to control ($p < 0.01$). When either MSH/ACTH 4-10 or physostigmine were administered in conjunction with atropine, DRP was found not to significantly differ from control. As previously stated, MSH/ACTH 4-10 at a dose of 95 ug/kg resulted in DRP significantly better than control ($p < 0.05$). Physostigmine sulfate administration alone did not significantly alter DRP when compared to control. In comparison to MSH/ACTH peptide and physostigmine administration, neither strychnine nor methylphenidate injection resulted in the reversal of cognitive impairment of atropine. Both of the latter in combination with atropine resulted in impaired DRP when compared to control ($p < 0.01$). Strychnine sulfate, but not methylphenidate HC1, when given alone resulted in a significant improvement in DRP when compared to control ($p < 0.05$).

Similar to the results obtained after atropine administration, the combination of either MSH/ACTH 4-10 95 ug/kg or physostigmine sulfate 0.5 mg/kg with imipramine HC1 10.0 mg/kg resulted in DRP not differing from control. Thus, both MSH/ACTH peptide and physostigmine administration can functionally reverse the anticholinergic-induced memory inpairments caused by either atropine sulfate or imipramine HC1.

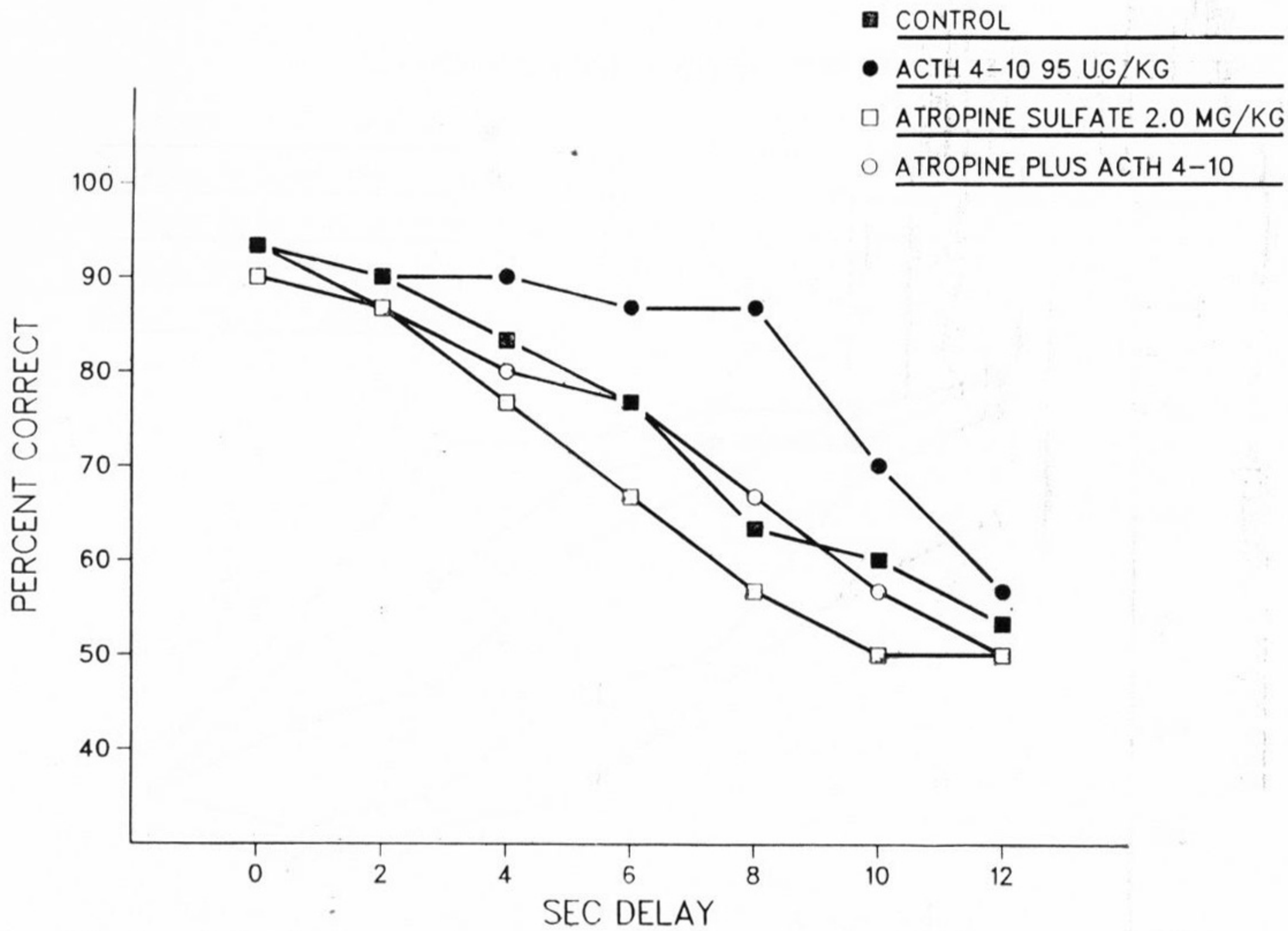


Fig. 3. — *Delayed response performance after atropine sulfate 2.0 mg/kg treatment, alone* **and in conjunction with MSH/ACTH 4-10 95 ug/kg. Data expressed as mean percent correct** *at each delay period.*

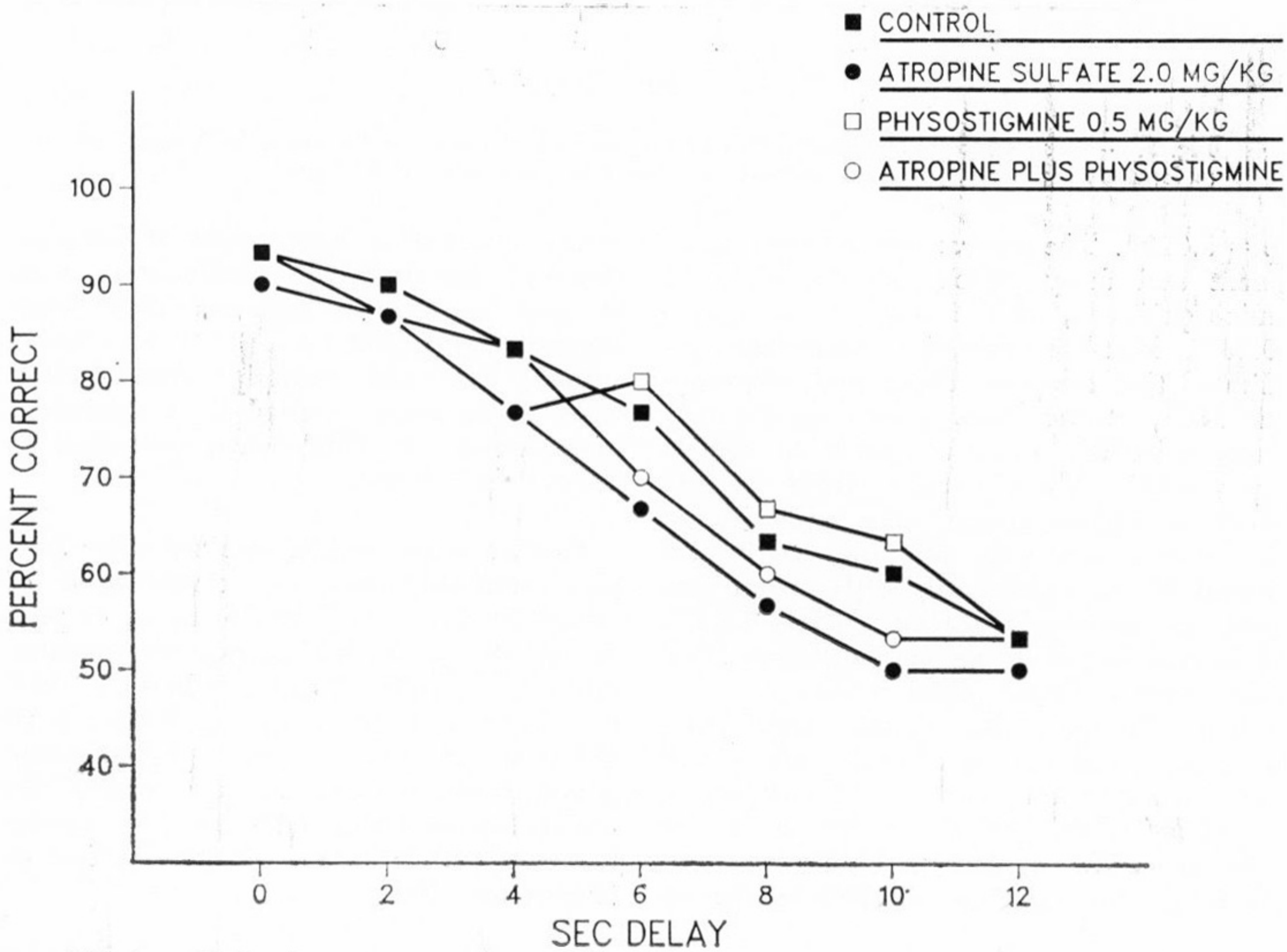


Fig. 4. — *Delayed response performance after atropine sulfate 2.0 mg/kg treatment, alone and in conjuncion with physostigmine sulfate 0.5 mg/kg. Data expressed as mean percent correct at each delay period.*

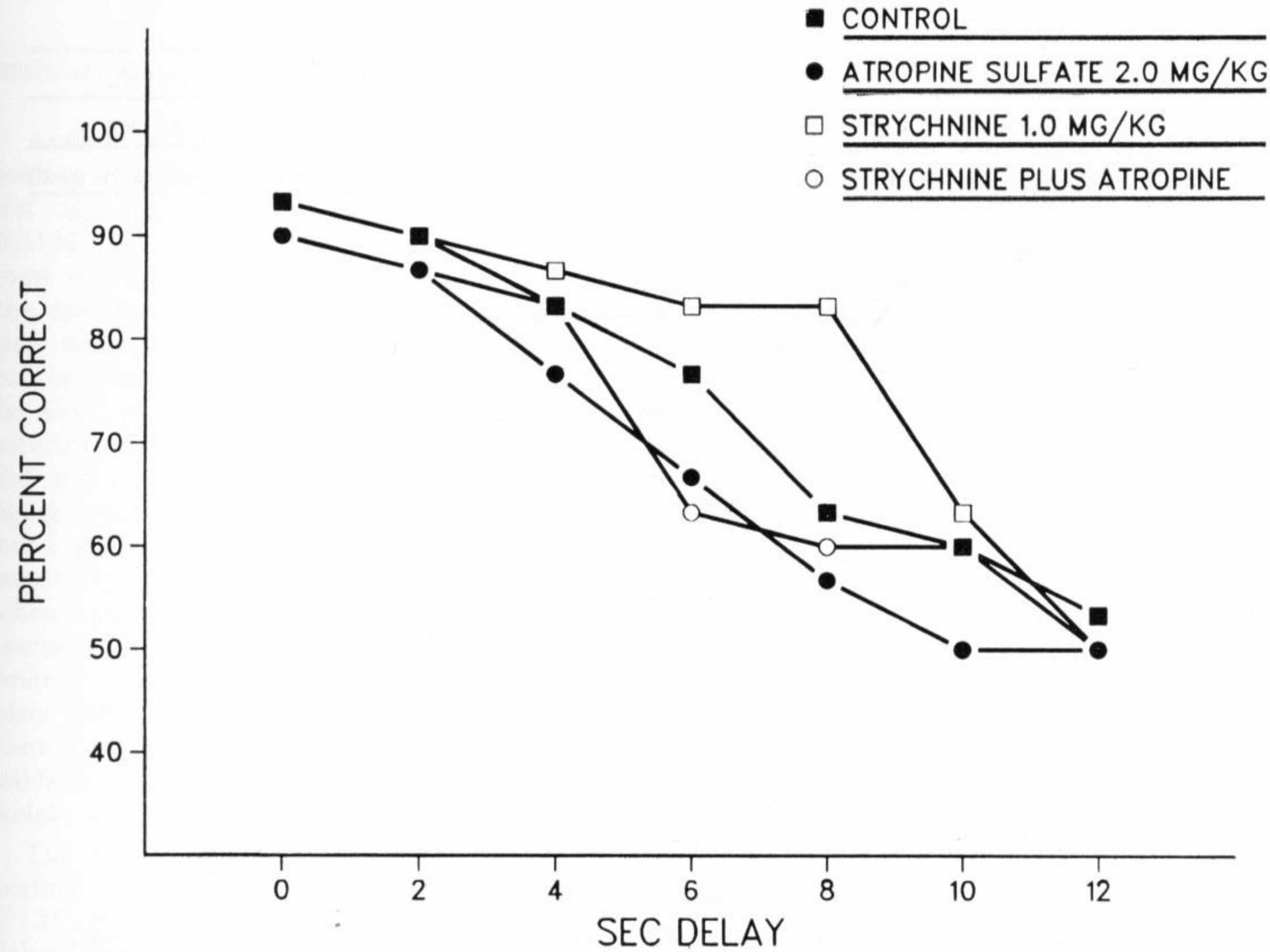


Fig. 5. — *Delayed response performance after atropine sulfate 2.0 mg/kg treatment, alone and in conjunction with strychnine sulfate 1.0 mg/kg. Data expressed as mean percent correct at each delay period.*

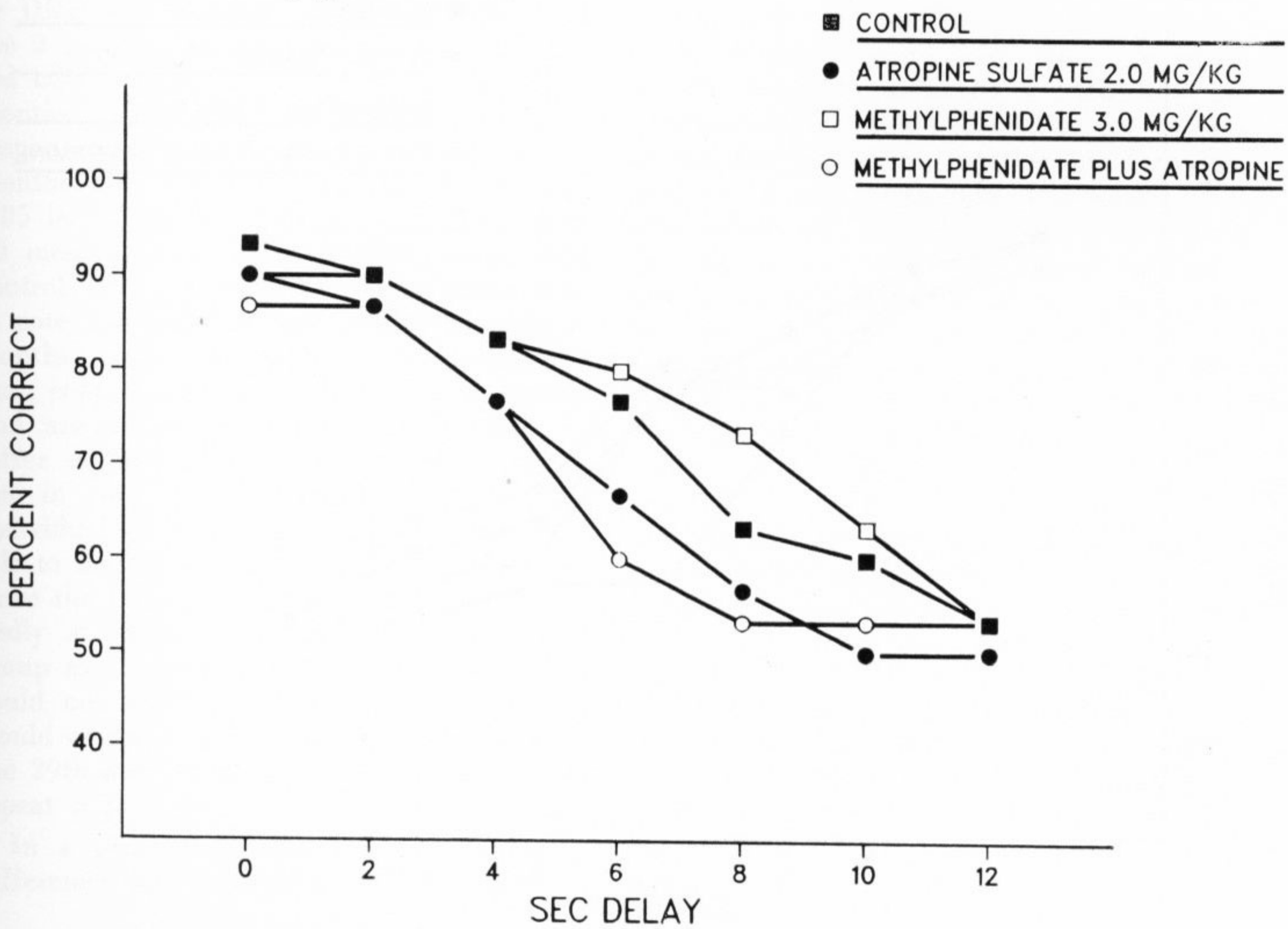


Fig. 6. — *Delayed response performance after atropine sulfate 2.0 mg/kg treatment, alone and in conjuncion with methylphenidate hydrochloride 0.E mg/kg. Data expressed as mean percent correct at each delay period.*

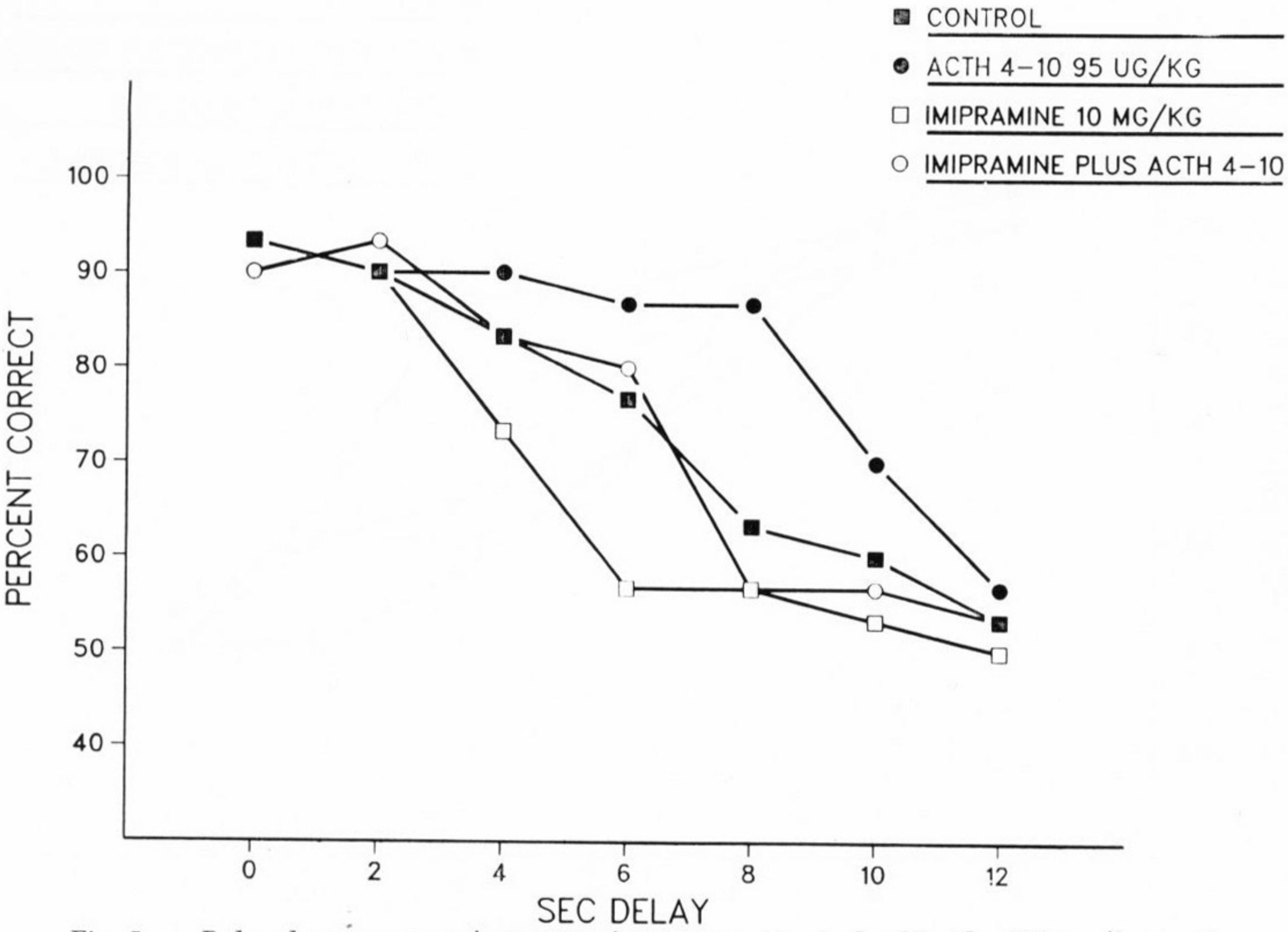


Fig. 7. — *Delayed response performance after imipramine hydrochloride 10.0 mg/kg treatment, alone and in conjunction with MSH/ACTH 4-10 95 ug/kg. Data expressed as mean percent correct at each delay period.*

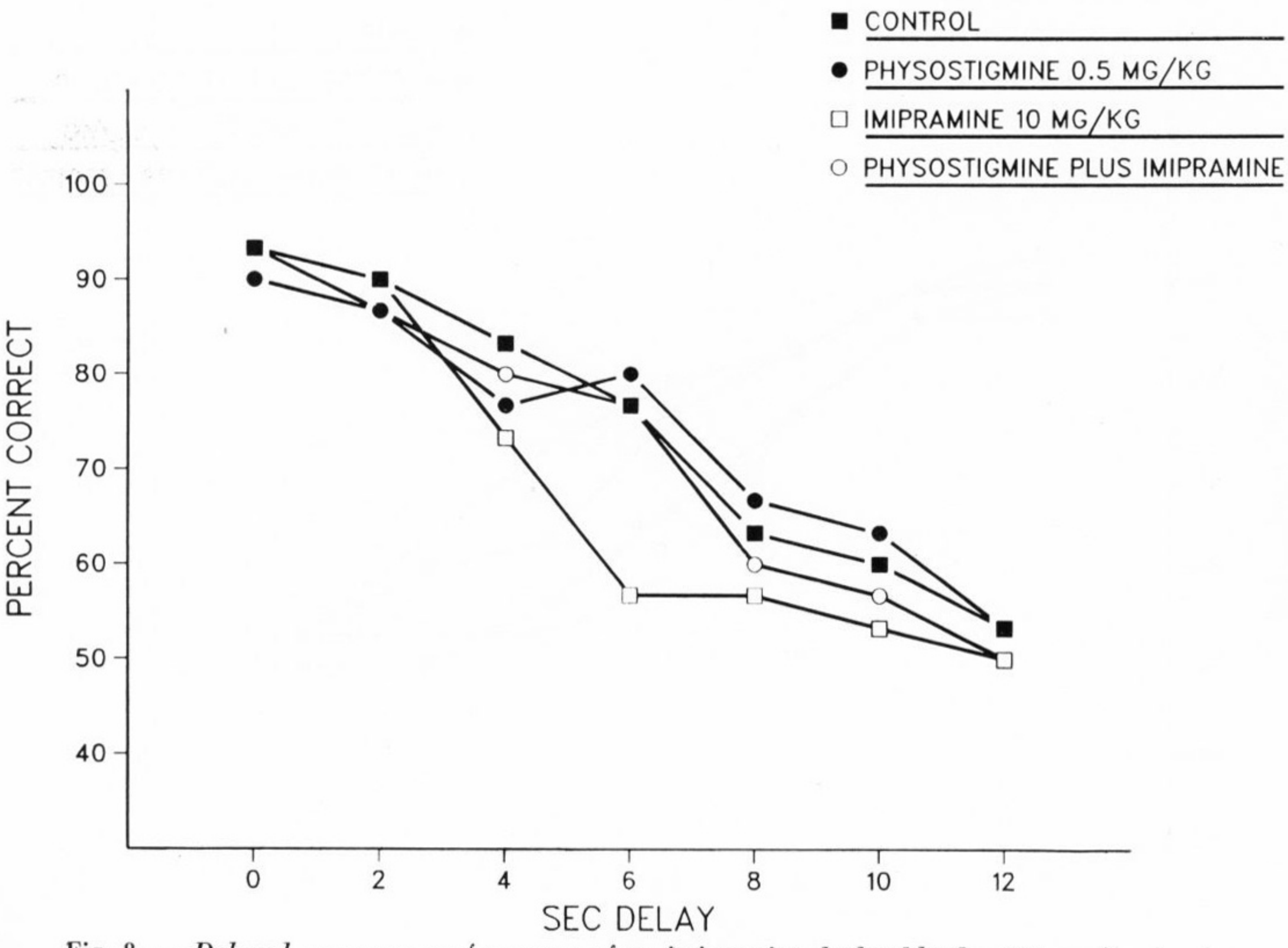


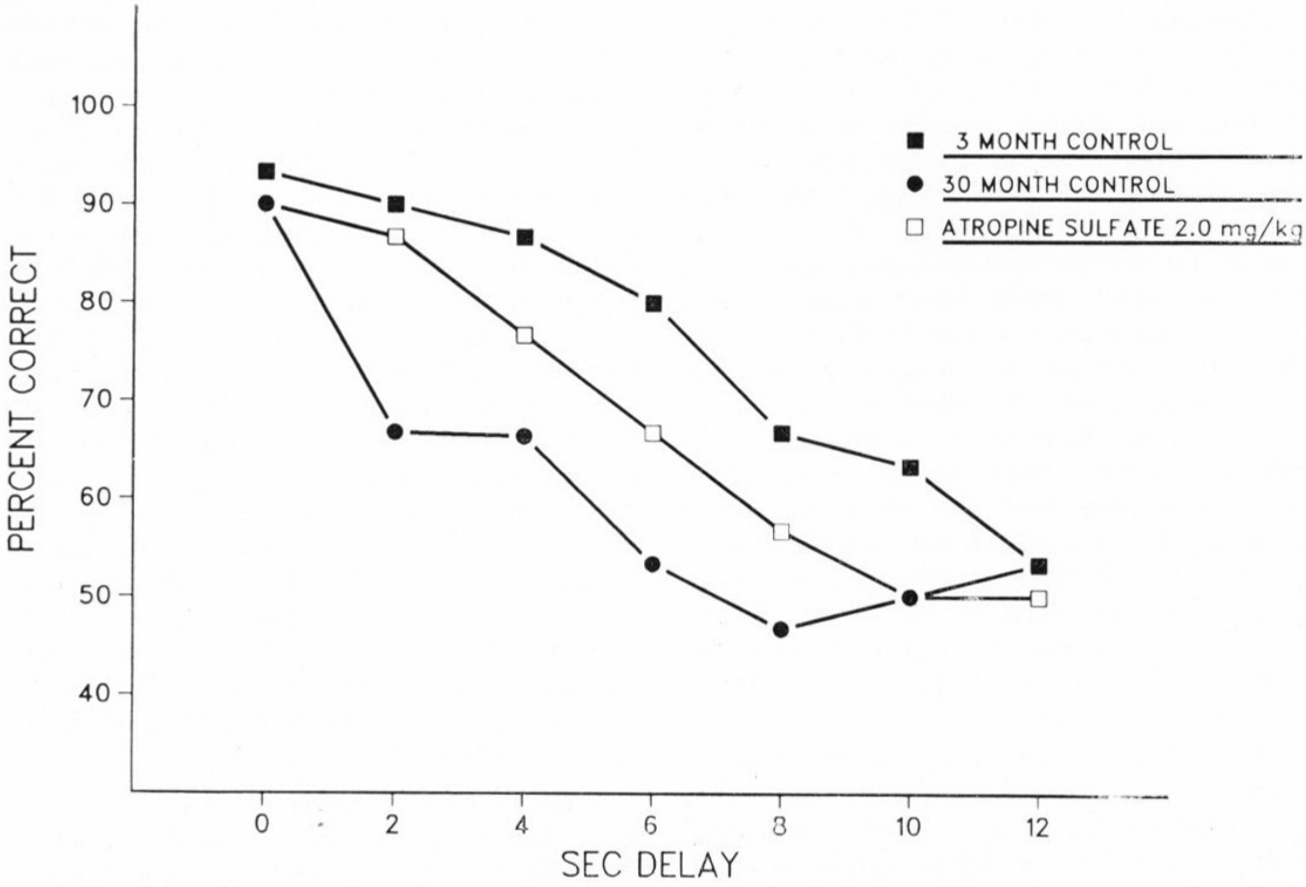
Fig. 8. — *Delayed response performance after imipramine hydrochlorde 10.0 mg/kg treatment, alone and in conjunction with physostigmine sulfate 0.5 mg/kg. Data expressed as mean percent correct at each delay interval.*

Analysis of variance of the animals' free feeding weights as aging progressed was not significant, $F(6,30) = 0.49$; $p = 0.8108$. Ad libitum weights remained almost constant throughout the study as did the calculated 85 percent free feeding testing weights of the animals. From this it can be interpreted that changing motivation for food, which would have resulted in significant alterations in free feeding weights, did not contribute to changes in DRP as aging progressed. In addition, the continued partial deprivation of food did not result in larger than normal food intake when unlimited food was available. Initial increases in food intake may have occurred prior to free feeding weighings which took place after one week of free access to lab chow. Early increases in weight were possible, and probable, before ad libitum weight determinations.

The effect of age on delayed response performance proved to be significant, $F(21,105) = 2.08$; $p = 0.0081$. The age/delay interaction was found not to be significant, $F(126,630) = 1.13$; $p = 0.1750$. Post-hoc analyses yielded several ages where DRP was notably worse than DRP at the 3 month baseline. Significantly impaired DRP was not found until an age of 23 months ($p < 0.05$). In addition, delayed response performance at ages of 25 and 26 months also differed from control at the 0.05 level. The last two ages tested, 29 and 30 months, were more notably worse that control DRP ($p < 0.01$). It is interesting to note that DRP at ages of 24, 27 and 28 months were not significantly different than control although DRP at ages among and around these months were found to differ significantly from control. This is due in part to the variability of DRP in individual animals which caused group totals to drift near the level of significance. Once the DRP of one animal dropped markedly at one month, which caused the group as a whole to differ from control, it could not be stated that particular animal would continue to perform poorly. Only at the 29th and 30th month did most animals repeat a poor performance.

In a planned comparison, a significant difference was found between DRP in young control animals, 30 month controls and animals after atropine administration, $F(2,10) = 16.23$; $p = 0.0007$. The treatment/delay interaction was not significant. Post-hoc analysis revealed that animals when aged demonstrated significantly worse DRP than after atropine administration when young ($p < 0.05$). As expected, DRP in young controls differed from that after atropine injection ($p < 0.05$), and young controls performed significantly better than when aged ($p < 0.01$). While DRP of young animals given atropine sulfate 2.0 mg/kg and animals when aged are both significantly worse than young control performance levels, aging results in the significantly poorest DRP of the three. If one were to sequentially compare animal DRP at each progressive age, it is certain that one would find DRP at one or more ages that did not differ significantly from DRP after atropine administration.

Analysis of the age-related MSH/ACTH 4-10 dose response data yielded a significant effect of age on DRP, $F(1,5) = 43.90$; $p = 0.0012$. Treatment condition also was found to be significant, $F(4,20) = 4.51$; $p = 0.0092$. In addition, several interactions reached significance. The age/treatment interaction achieved a high level of significance, $F(4,20) = 11.73$; $p < 0.0001$, as did the age/delay interaction, $F(6,30) = 4.12$; $p = 0.0039$. The treatment/delay interaction was not significant, $F(24,120) = 0.92$; $p = 0.5720$. The three-way interaction of age/treatment/delay approached but did not achieve significance, $F(24,120) = 1.56$; $p = 0.0613$. Post-hoc analyses yielded several significances when treatments, young versus aged, were compared. Control at 3 months was found to have better DRP when compared to 30 month vehicle ($p < 0.05$). MSH/ACTH 4-10 ug/kg, as well as 95 ug/kg treatment, yielded significantly beter DRP at 3 months than at 30 months of age ($p < 0.05$). Administration of MSH/ACTH 4-10 195 ug/kg at 3 and 30 months did not result in differing DRP when compared to each other. However, MSH/ACTH peptide administration at a dose of 285 ug/kg resulted in significantly better DRP in aged animals than when young ($p < 0.05$).



Fgi. 9. — *Delayed response performance at age 3 months, age 30 months and after atropine sulfate 2.0 mg/kg administration. Data expressed as mean percent correct at each period of delay.*

In young animals MSH/ACTH 4-10 95 ug/kg administration resulted in significantly better DRP than 3 month control ($p < 0.05$). The two higher doses, 195 and 285 ug/kg, yielded significantly worse DRP than the respective control ($p<0.05$). MSH/ACTH peptide administration in older animals yielded significantly better DRP than 30 month control in all but the lowest dose. MSH/ACTH 4-10 95 ug/kg and 195 ug/kg resulted in DRP significantly better than older control at the 0.05 level, while the highest dose of the peptide yielded better DRP than control at the 0.01 level.

The age/delay treatment interaction also yielded several post-hoc significances. Animals at 3 months of age performed significantly better than when aged at 6 second delay (77 vs 54 percent correct, $p<0.05$), 8 second delay (64 vs 46 percent correct, $p<0.05$) and 10 seconds of delay (60 vs 50 percent correct, $p<0.05$).

Trend analysis for the MSH/ACTH peptide dose responses, young and old, yielded several significant trend components. Younger animals were found to have significant linear, $F(1,120) = 10.1$; $p < 0.01$, and quadratic, $F(1,120) = 6.5$; $p < 0.025$, components. The cubic and quartic trend components were not significant for 3 month old animals. In contrast, older animals had only a significant linear trend, $F(1,120) = 4.6$, $p < 0.05$. This can be visualized in Figure 11.

When animal age is correlated with individual DRP a significant negative relationship is found, $r = -0.455$, $p < 0.0001$. When mean performance is correlated with age a stronger relationship is obtained, $r = -0.847$, $p < 0.0001$. Thus, as age increases DRP decreases in a significantly associated fashion. Regression equations for individual and average DRP are extremely similar but not exact. For individual DRP results correlated with age, a Y intercept

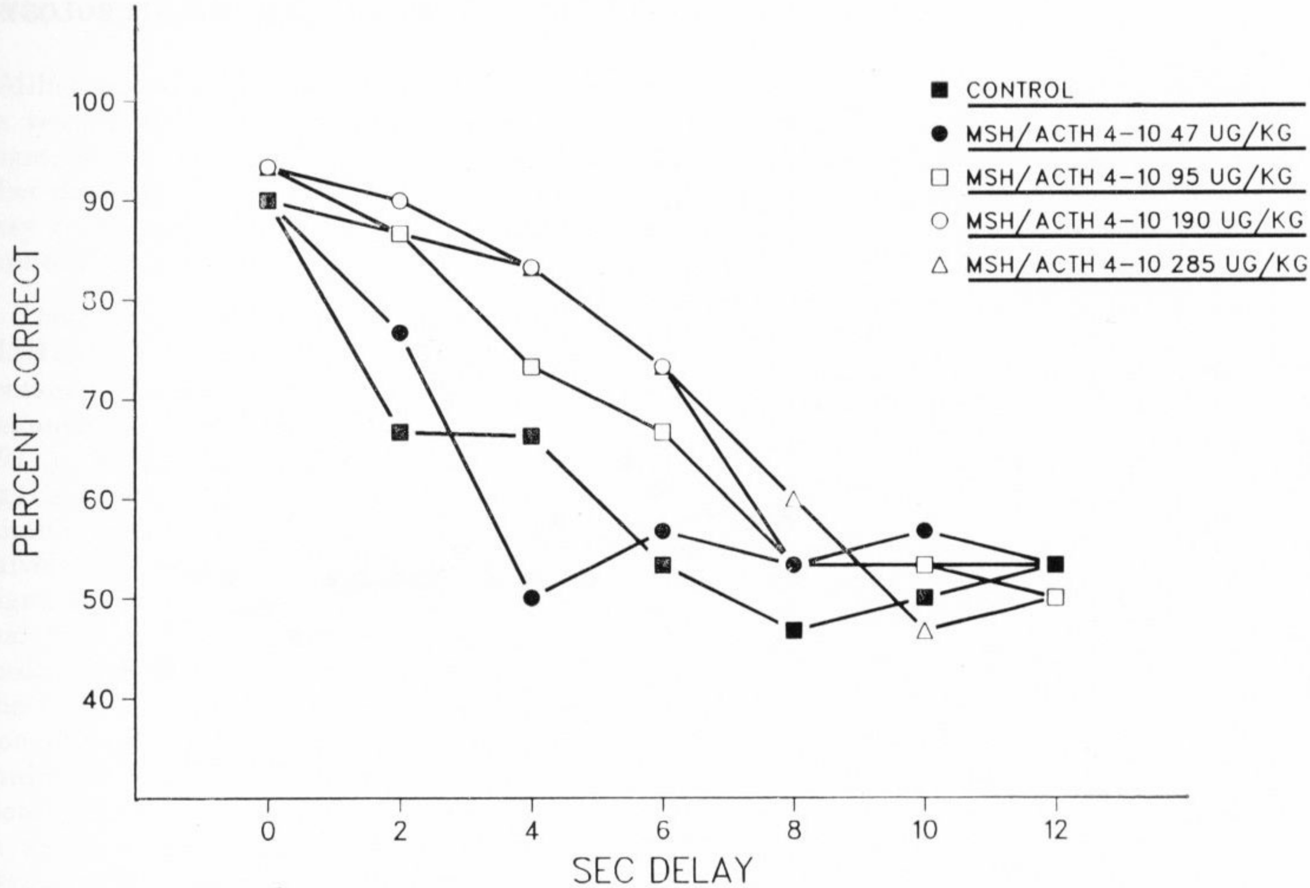


Fig. 10. — *MSH/ACTH 4-10 dose response on delayed response performance in aged animals. Data expressed as mean percent correct at each delay period.*

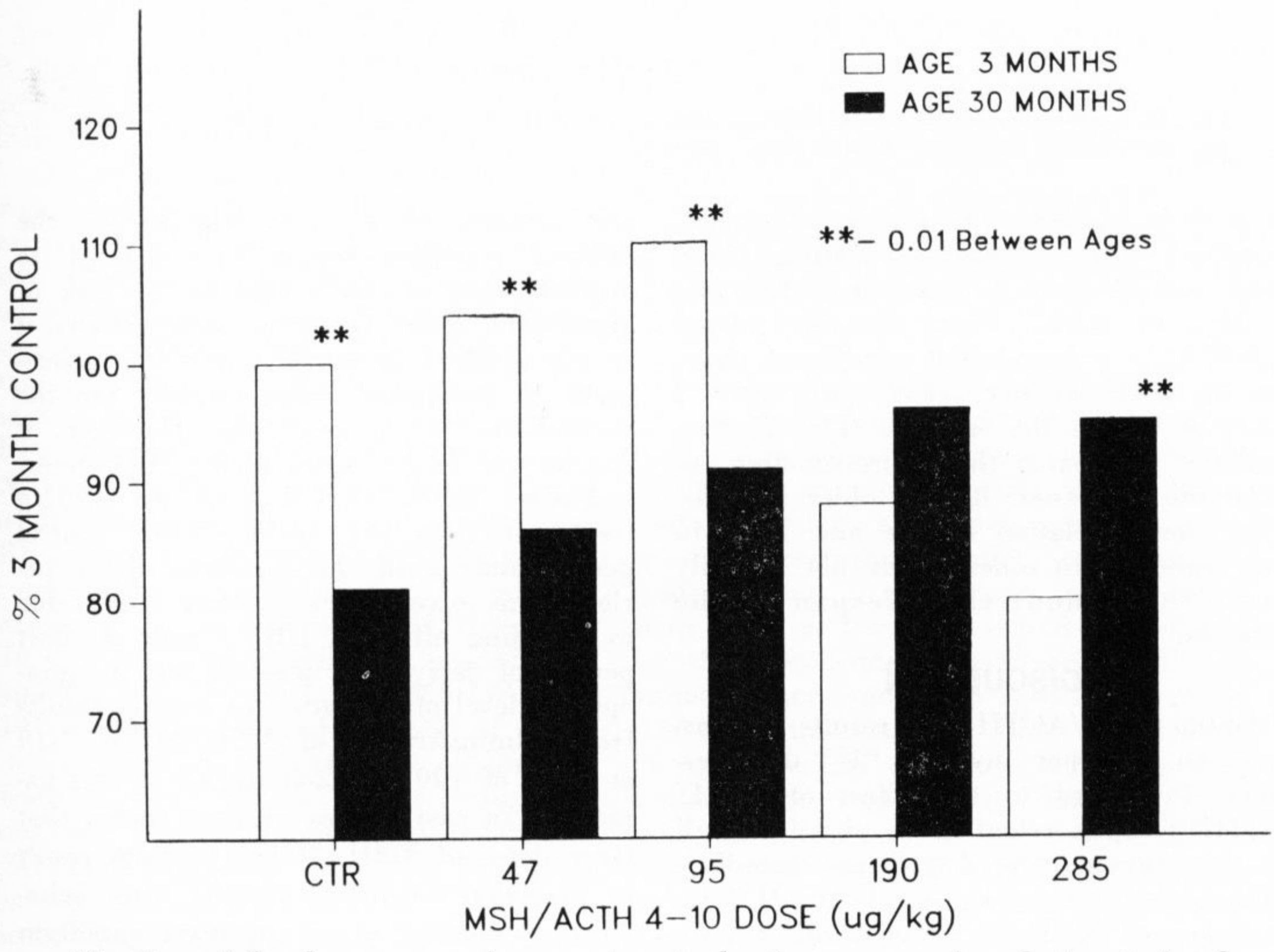


Fig. 11. — *Delayed response performance in animals when young and aged at control and each dose of MSH/ACTH 4-10 tested. Data expressed as mean percent correct collapsed across delay periods.*

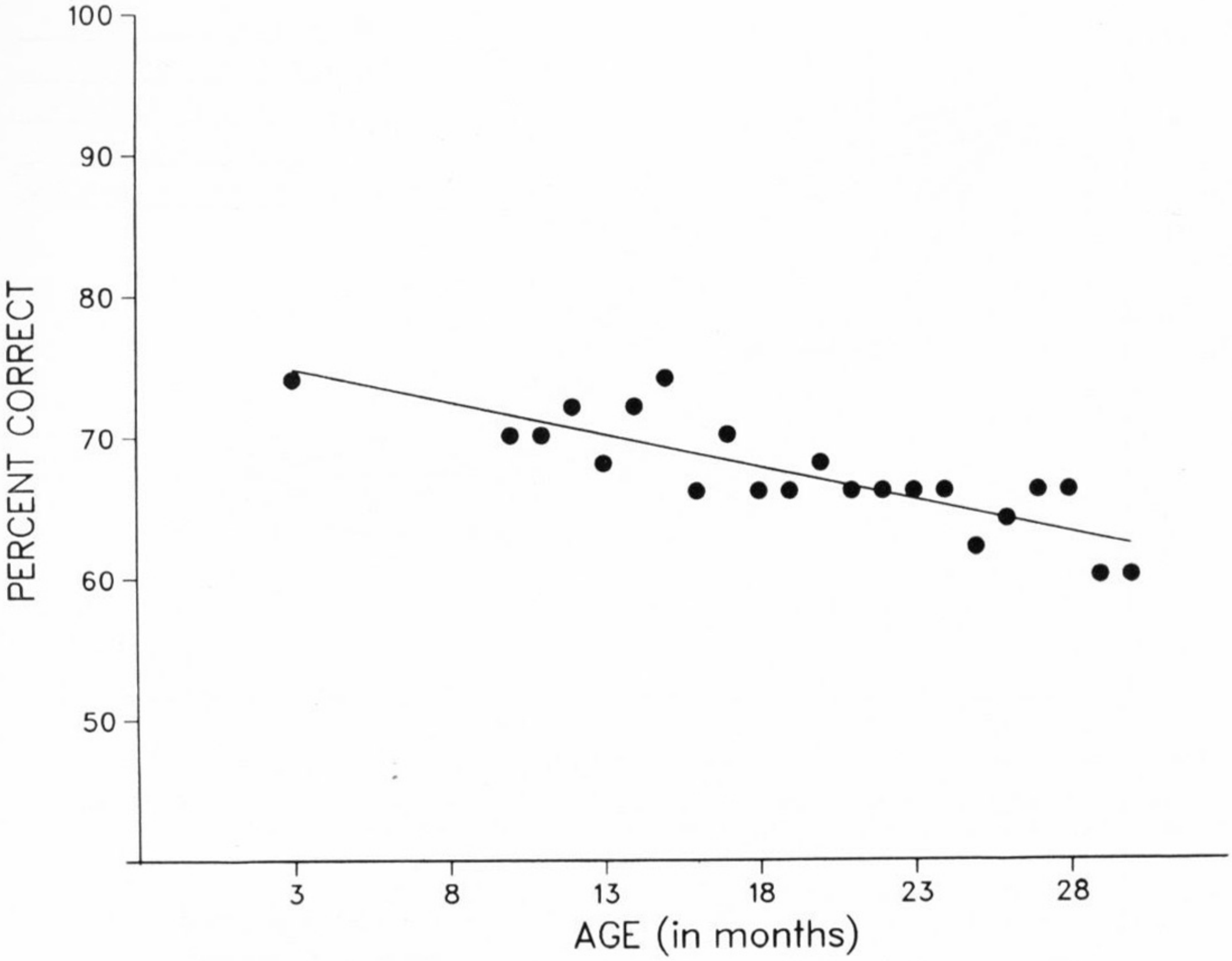


Fig. 12. — *Regression for mean DRP at ages of 3 and 10-30 months. Data expressed as mean percent correct collapsed across delay periods.*

of 3.79 is yielded with a slope of -0.022. Obtained results from correlating mean DRP and age are a Y intercept of 3.81 and a slope of -0.023. From the age related ANOVA, it is found that significant changes in DRP do not occur until after 2 years of age in the subjects. This finding explains, in part, the overestimation of DRP the regression line in older animals. The true correlation of age and DRP in this paradigm in rodents may not be truly linear throughout the lifespan of the subjects.

DISCUSSION

Initial MSH/ACTH 4-10 results demonstrate an apparent inverted "U" dose response in regard to the effect of MSH/ACTH 4-10 on retention of visual stimuli in the Hunter delayed response paradigm in animals when young. MSH/ACTH 4-10, at a dose of 95 ug/kg, significantly improved memory of a visual stimulus in the delayed response apparatus compared to animals' control DRP. Due to the lack of significant delay/treatment interactions it is not prudent to perform post-hoc assessment at individual delay periods for the various treatment conditions. However, it can be seen from Figure 2 that DRP observed after MSH/ACTH 4-10 95 ug/kg treatment does not deviate from control performance until the 4 second delay period. This observation is most likely due to a ceiling effect of DRP found at short periods of delay. It is possible that a supra-optimal level of excitation or arousal results from administration of MSH/ACTH 4-10 at doses of 190 and 285 ug/kg in this paradigm. In past studies we have shown that large doses of MSH/ACTH peptides result in increased locomotor activity after behavioral assessment in an appetitive paradigm

(Miller and Turnbull; 1986). A reduction in reward attainment in the Hunter paradigm, which was noted when the two higher doses of MSH/ACTH 4-10 were given, may contribute to this observed increase in post-test locomotion.

These results support the concept of MSH/ACTH peptide enhancement of attentional processes originally postulated by Sandman and co-workers (Sandman, et al., 1977). MSH/ACTH 4-10 at a dose of 95 ug/kg clearly enhanced memory of visual stimuli as measured by DRP in an appetitively motivated delayed response paradigm. It can be hypothesized that an optimal level of attention or excitation was exceeded by administration of the two highest doses of the peptide. The demonstration of an inverted "U" dose response regarding a MSH/ACTH peptide and behavioral performance in animals when young, in an appetitive paradigm points out the necessity for the use of various doses of neuropeptides in assessing their ability to alter behavior.

Atropine and imipramine administered IP significantly impair DRP, presumably due to their anticholinergic effects. When MSH/ACTH 4-10 is given IP, at a dose of 95 ug/kg, in conjunction with either atropine or imipramine, DRP returns to control vehicle levels. A similar effect is noted when physostigmine is given in conjunction with either anticholinergic agents. However, neither methylphenidate nor strychnine have any effect on atropine-induced cognitive deficits as measured by DRP. The specificity of physostigmine and MSH/ACTH 4-10 in the reversal of muscarinic blockade-induced impairment of DRP as opposed to either methylphenidate or strychnine supports direct cholinergic involvement in the effect of MSH/ACTH 4-10 and atropine on DRP. A possible explanation for the effect of MSH/ACTH 4-10 on atropine-induced decrease of DRP is a trans-synaptic regulation of cholinergic release in the CNS by corticotropin. Enhancement of cholinergic transmission by MSH/ACTH peptides has been found to exist at the neuromuscular junction (Strand and Smith, 1980). From their studies and the work of others (Johnston, et al., 1983), it is evident that the MSH/ACTH peptides affect motor neurons at the cell body in the spinal cord causing increased release of ACh onto the muscle. Thus, a neuron in the CNS has been shown to increase ACh release upon MSH/ACTH peptide administration. While such a mechanism has also been proposed for septohippocampal cholinergic neurons, a negative finding was reported for the effect of MSH/ACTH 4-10 on ACh concentration in nerve terminals in the dorsal hippocampus (Botticelli and Wurtman, 1982). Assay of such small quantities of neurotransmitter is extremely difficult with large variance in findings possibly negating small, yet biologically significant alterations in ACh release.

Based on the present results, the behavioral effects of MSH/ACTH 4-10 on delayed response performance in the Hunter paradigm may be mediated, at least in part, by enhancement of the cholinergic system(s) in the CNS.

The specific reversal of atropine-induced impairments in DRP by MSH/ACTH 4-10 may represent functional evidence for the mediation of the behavioral effects of opiomelanocortin peptides by the cholinergic neuronal system. If true, agerelated changes in ACh levels in the CNS would lead to the expectation of different MSH/ACTH 4-10 dose-response profiles on DRP in young and aged rodents.

Following of DRP throughout the life span of the rodent subjects yielded interesting information in regard to age-related changes in cognitive performance. While confounds such as changes in sensory acuity, motor performance and food preference cannot be totally ignored, the assessment of short delay performance served as a control for these problems. In comparing DRP in animals when young and aged it was found that the animals did not differ significantly in early periods of delay. While animals demonstrated significantly impaired DRP as aging progressed, an age at which DRP became and remained significantly impaired can not be stated. Animals performed poorly one month, then similar to 3 month baseline the next. Individual variablility from month to month increased dramatically as animals aged. Perhaps this finding is more important than the occasional observed significant impairment in DRP which was found to occur in aging animals.

The two dose response curves obtained for MSH/ACTH 4-10 in the delayed response apparatus (young and aged) may possibly be interpreted in terms of the age related changes in the ACh system. Examination of MSH/ACTH 4-10 dose-response curves reveals an inverted "U" relationship for animals when young and a positive, more linear relationship for animals when aged.

One possible explanation for the differential dose-responses of MSH/ACTH 4-10 found between young and aged animals in the Hunter paradigm is the functional age-induced decrease in acetylcholine levels found in aged animals. Decreases in endogenous ACh levels may account for the observation that older animals do not demonstrate the observed impairment of DRP with higher doses of the MSH/ACTH peptide that is found in animals when younger. Possible hypersecretion of ACh in the CNS, and resulting DRP impairment, in response to high doses of MSH/ACTH 4-10 may not be found in animals when older due to the decreased amounts of ACh found in neural tissue due to the aging process. The present findings, taken with past work implicating enhancement of cholinergic transmission in the mediation of cognitive effects of MSH/ACTH peptide fragments, support the concept that age-induced decreases in neuronal acetylcholine levels may result in differential MSH/ACTH fragment dose-response profiles between young and aged animals in cognitive tasks.

Initially, it is hoped that the work of Bartus and co-workers which depict cholinergic blockade-induced alterations in cognitive performance similar to age-induced changes in memory, can be reproduced in rodents. However, DRP in animals at age 30 months proved to be significantly worse than animals when young after administration of atropine sulfate 2.0 mg/kg. In retrospect, this problem was built into the design of the study, for doses of drugs chosen in pilot examinations were those having significant yet minimal effects on DRP. Assessment of an atropine dose response in animals when young in the Hunter paradigm would be desirable and would have possibly eliminated the problem of aged versus anticholinergic DRP comparison.

All findings in the present study can be explained in terms of modulation of the ACh system. While further biochemical work is needed to validate the proposed ACh mediation of the behavioral actions of MSH/ACTH peptides, this work constitutes the first behavioral evidence suggesting increased ACh release in response to MSH/ACTH 4-10 administration. The clinical implications as to the treatment of age-induced or Alzheimer's related cognitive pathologies, which seem to be cholinergically mediated (Davis, et al., 1982), are obvious. The ACh precursor loading therapy and/or ACh uptake facilitation therapy could be reinforced with MSH/ACTH peptide or analog administration. Separately, these interventions have had mixed success; together the effect may be more substantial.

SUMMARY

The present study examines the effect of MSH/ACTH 4-10 on delayed response performance (DRP) in a two choice version of the Hunter paradigm incorporating the Honzik opaque door modification. DRP was measured in male, Long-Evans rats after administration (IP) of various doses of MSH/ACTH 4-10 or control when animals were young (3 months) and aged (30 months) and aged (30 months). Between MSH/ACTH dose response observations animals received a variety of psychoactive agents of various classes prior to DRP assessment. In addition, DRP was assessed once per month, without drug, from ages 10 to 30 months.

MSH/ACTH 4-10, at a dose of 95 ug/kg, significantly enhanced retention of

a visual stimulus, while larger doses of MSH/ACTH 4-10 impaired delayed response performance in animals when young. Reversal of anticholinergic-induced DRP impairments by physostigmine and MSH/ACTH 4-10, but not strychnine or methylphenidate, suggests that the effect of MSH/ACTH 4-10 on DRP is specific and may be mediated by enhancement of the cholinergic system(s) in the CNS.

Animals began to demonstrate significant impairment in DRP, at longer delays, at the age of 23 months. While confounds such as changes in sensory acuity, motor performance and food preferance cannot be totally ignored, the assessment of performance at shorter delay periods served as a control for these problems. No significant age-related changes in DRP at shorter periods of delay were found, indicating that perception and motor capabilities played little role in age-related DRP alterations.

Trend analysis revealed that animals demonstrate significant linear and quadratic MSH/ACTH 4-10 dose responses which appear as an inverted "U" in the Hunter paradigm when young. As animals age, this dose response becomes a purely positive linear relationship. Thus, the age-induced decrease in acetylcholine (ACh) levels, and hence MSH/ACTH peptide-induced release, may result in the change in MSH/ACTH dose response profiles. These findings may have clinical implications in the treatment of age-induced or Alzheimer's related cognitive pathologies, which are of cholinergic etiology.

RESUMEN

El Presente estudio examina el efecto de MSH/ACTH 4-10 sobre la ejecución de respuesta de morada (DRP) en una versión de doble elección del paradigna de Hunter incorporando la modificación de puerta opaca de Honzik. DRP fue valorado en ratas machos Long-Evans después de administración de varias dosis de MSH/ACTH 4-10 o control cuando los animales eran jóvenes (3 meses) y mayores (30 meses). Entre observaciones de respuesta a la dosis MSH/ACTH, los animales recibieron una variedad de agentes psicoactivos de varias clases antes de la valoración del DRP (Delayed Response Performance). Además DRP fue apreciado una vez por mes, sin drogas, de edades de 10 a 30 meses.

MSH/ACTH 4-10 a una dosis de 95 ug/kg, significativamente acrecienta la terción de un estímulo visual, mientras que dosis mayores de MSH/ACTH 4-10 afecta la ejecución de respuesta demorada en animales jóvenes. Revocación de dificultades inducidas en DRP por anticolinérgicos por fisostigmina y MSH/ACTH 4-10 pero no por estrignina o metilfenidate, sugiere que el efecto de MSH/ACTH 4-10 sobre DRP es específico y puede ser mediado por acrecentarse el sistema colinérgico en el sistema nervioso central.

Los animales comienzan a demostrar significativos defectos en DRP A mayor dilución a la edad de 23 meses. Mientras confusiones tales como cambios en la agudeza sensitiva, ejecución motora y realización de alimentos no puede ser totalmente ignoradas la comprobación de ejecución a períodos de demora más cortos sirvió como control para estos problemas. No fueron encontrados cambios significativos en relación con la edad en DRP en más breves períodos de demora indicando que la percepción y capacitación motora juegan un rol discreto en alteraciones de DRP relacionadas con la edad.

Análisis de dirección reveló que los animales demostraron significativas respuestas lineal y cuadrática a la dosis de MSH/ACTH 4-10 lo caul aparece como una "U" invertida en el paradigma de Hunter cuando joven. En lo relativo a la edad del animal esta respuesta la dosis llega a ser una relación lineal puramente positiva. De este modo el decrecimiento inducido por la edad en niveles de acetilcolina (ACH) y de liberación MSH/ACTH péptida inducida, puede resultar en el cambio en perfiles de respuesta por dosis MSH/ACTH. Estos hallazgos pueden tener implicaciones clínicas en el tratamiento en patológicos cognitivas inducidas por la edad o el Alzheimer las cuales son de etiología colinérgica.

RÉSUMÉ

Cette étude s'applique à l'action du MSH/ACTH 4-10 sur la performance de la réponse différée (PRD) dans deux versions du paradigme de Hunter comprenant une modification de la porte opaque de Honzik. La PRD a été mesurée sur des rats mâles Long-Evans après l'administration de doses variées de MSH/ACTH 4-10 ou d'un contrôle, alors que les animaux étaient jeunes (3 mois) et âgés (30 mois). Entre les observations des réponses à la dose de MSH/ACTH, les animaux recevaient divers agents psychoactifs avant la détermination de la PRD. D'autre part, la PRD était mesurée une fois par mois du dixième au trentième mois, sans médication.

MSH/ACTH 4-10, à la dose de 95 ug/kg, éléve de façon significative la rétention d'un stimulus visuel, alors que des doses plus élevées abaissent les performances chez les jeunes animaux. L'abaissement de la PRD produite par les anticholinergiques est annulé par la physiostigmine et par le MSH/ACTH 4-10, mais non par la strychnine ni le méthylphénidate. Ceci suggére que l'action du MSH/ACTH 4-10 sur la PRD est spécifique et peut être transmise par la mise en action du (ou des) système cholinergique au niveau du système nerveux central.

A plus long délai, les animaux commencent à montrer une diminution significative de la PRD à l'âge de 23 mois. La confusion possible entre de telles modifications de l'acuité sensorielle, les performances motrices et les préférences alimentaires ne peut être ignorée et la détermination de la performance à de plus courts délais en permet le contrôle. Le fait qu'on n'ait pas trouvé de modifications significatives de la PRD liées à l'âge aux contrôles à plus courts délais indique que les possibilités perceptives et motrices ne jouent qu'un faible rôle dans les altérations du PRD rapportées à l'âge.

Pour le paradigme de Hunter, les réponses aux doses de MSH/ACTH, linéaires ou carrées, se présentent sous forme d'un U renversé lorsque les animaux sont jeunes. Chez l'animal plus âgé, la réponse devient purement linéaire. Ainsi, la diminution avec l'âge des taux d'acétylcholine et la libération de MSH/ACTH par induction peptidique, peut aboutir à la modification des profils de la réponse aux doses de MSH/ACTH. Ces constatations peuvent avoir des implications cliniques dans le traitement des pathologies cognitives en relation avec l'âge ou avec la maladie d'Alzheimer, qui sont d'étiologie cholinergique.

ZUSAMMENFASSUNG

In vorliegender Arbeit wird die Wirkung von MSH/ACTH 4-10 auf die Spaete Reaktive Realisierung (DRP) untersucht, bei einer Version doppelter Wahl des Huntey Paradigmas mit der Modifikation der Dunklen Tuer von Honzig. DRP wurde bei maennlichen Long Evanz Ratten gemessen nach der Verfuetterung verschiedener Dosen von MSH/ACTH 4-10 oder bei Kontrolltieren, wenn diese sehr jung (9 monate) oder sehr alt (30 Monate) waren. Bei den Beobachtungen der Reaktion zwischen den MSH/ACTH Verfuetterungen erhielten die Tiere verschiedene psychoactive Mittel unterschiedlicher Art vor der DRP -Messung. Ausserden wurde die DRP einmal im Monat ohne Medikamente durchgefuehrt bei Tiern zwischen 10 und 30 Monaten.

MSH/ACTH 4-10 bei einer Dosis von 95 microgtamm/kg verstaerkte bedeutend die Retention des visuellen Reizes, waehrend grosse Dosen von MSH/ACTH 4-10 bei jungen Tieren die DRP verschlechterten. Die Annulierung der Verschlechterung der durch Anticholinergische Drogen induzierten DRP wurde durch Physostigmin und MSH/ACTH 4-10, aber nicht durch Strychnin oder Methylphenidat herbeigefuehrt. Was bedeuten kann, dass die Wirkung von MSH/ACTH auf die DRP spezifisch ist und wohl durch die Verstaerkung des cholinergische Systems im Zentralnervensystem vermittelt wird.

Die Tiere begannen eine bedeutende Verschlechterung der DRP zu zeigen bei laengeren Verzoegerungen im Alter von 23 Monaten. Waehrend Verschlechterungen.

wie der Sensibilitaetsschaerfe, der motorischen Beweglichkeit und der Nahrungswahl nicht vollkommen ignoriert werden koennen, diente die Pruefung der Leistungenfen nach kuerzerdauernden Verzoegerungen als Kontrolle dieser Probleme. Man fand keine signifikanten altersbedigten Veraenderungen der DRP bei kuerzer dauernden Verzoegerungen, was bedeuted, dass Perzeption und motorische Faehigkeiten eine geringe Rolle bei den altersbedingten Veraenderungen de DRP spielen.

Eine Analyse der Tendenzen zeigt, dass die Tiere bedeutende lineare und quadratische Reaktionen auf MSH/ACTH 4-10-Dosen aufwiesen und als umgekehrtes "U" im Hunterschen Paradigma erscheinen, wenn die Tiere jung sind. Wenn die Tiere aelter werden, erhaelt die Reaktion auf die Dosis ein klar positives liniaeres Verhaeltinis. So mag die altersbedingte Veraenderung des Azetylcholinspiegels (ACH), und infolgedessen, der durch Peptide verursachten Produktion von MSH/ACTH 4-10, die Aenderung der Reaktionsprofile auf die MSH/ACTH 4-10 Dosen verurssachen. Diese Befunde koennten klinische Bedeutung haben bei der Bahandlung der kognitiven krankhaften altersbelingten oder durch die Alzheimersche Krankheit verursachten Zustaende cholinergischer Aetiologie.

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Neurological and Behavioral Investigations of Memory Failure in Aging Animals

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Although there is a substantial degree of stability of certain mnemonic processes throughout the lifespan of healthy mammals, normal aged organisms do show changes in their ability to learn and remember under some conditions. It is important to emphasize that approximately 80 % of the human population will age "normally" and will not undergo the devastating neurological and cognitive declines sustained in syndromes such as Alzheimer's or multiinfarct dementias. The change in world demographic characteristics involving larger proportions of people aged 65 and over means that persons with dementing illness (about 20 % of the population in the United States) are placing increasingly greater demands on our health care systems. This makes the understanding of the etiology and underlying mechanisms of these diseases of primary medical and social importance. A basic understanding of normal aging is also of primary importance as it will provide the essential baseline against which all pathological conditions will need to be examined. An understanding of this process will also provide basic insights into the functions that are likely to be preserved and those that may provide problems for the great majority of the elderly population. As a number of speakers in this symposium have suggested, memory function is one aspect of both pathological and normal aging that is of great concern to older people themselves, as it is essential for their maintenance of independent and productive lives. In normal aging, the fear of failing memory is often worse than the actual demonstrated changes in the process. Nevertheless, there are certain alterations that do occur with age that are at least minimally annoying. If the basis for the selective changes in memory are discovered, then treatment strategies might be developed that would benefit the quality of life of a large segment of the population. This is the basic rationale for research programs using animal models of memory loss in aging, and has been the primary focus of the experiments from our laboratory that examine both behavioral memory and the electrophysiological functioning of brain structures thought to be critically involved in learning and memory processes.

To ensure that the animal model of normal aged human memory is appropriate, it is necessary to choose behaviors and memories that might find a correspondence between species, which of course leaves out language functions. For example, it appears that both aged humans and aged rats exhibit spatial memory deficits. It is more difficult for older organisms to learn about and to navigate accurately within a familiar or novel environment than it is for younger organisms (Barnes, 1979; de Toledo-Morrell and Morrell, 1985; Evans, Brennan, Skorpanich and Held, 1984; Gage, Dunnett and Bjorklund, 1984; Light and Zelinski, 1983; Ohata and Kirasic, 1983; Rapp, Rosenberg and Gallagher, 1987; Walsh, Krauss and Regnier, 1981; Weber, Brown and Weldon, 1978). This kind of spatial memory therefore provides an excellent point of convergence between species and is the type of memory that our laboratory has focused on in behavioral experiments with old animals. The other critical feature that recommends examining this type of memory is the fact that the neural structures that are critically involved in these behaviors are relatively well understood both in the rat and in humans. That is, the hippocampal formation is known to be very important for learning and memory processes (e.g., Scoville and Milner, 1957; Zola-Morgan, Squire and Amaral, 1986), which includes learning where one is in space. Temporal lobe resection, for example in Scoville and Milner's patient H.M., included a deficit in the way in which he found his way through his environment (Corkin, 1984). Lesions of this area in rats can also produce animals that do not utilize spatial information effectively (Jarrard, 1978; Morris, Garrud, Rawlins and O'Keefe, 1982; O'Keefe, Nadel, Keightly and Kill, 1975; Olton, Walker and Gage, 1978; Sutherland, Whishaw and Kolb, 1983). While this is not the only function that the hippocampus is likely to subserve, a correlation of spatial behavior with hippocampal physiology has been particularly useful in the delineation of some of the potential brain changes responsible for memory changes with age. The following is a selected review of the evidence, mostly from our laboratory, that hippocampal dysfunction in old age leads to deficits in spatial information processing.

The evidence that old rats possess spatial learning and memory problems has become very strong over the past decade. Old rats are worse at learning foraging problems on radial 8-arm mazes (Olton and Samuelson, 1976), where they must remember which of the 8 arms they have visited and have obtained reward from on a particular trial and which of the arms remain with food (e.g., Barnes, Nadel and Honig, 1980; Ingram, London and Goodrick, 1981; van Gool, Mirmiran and van Haaren, 1985; Wallace, Krauter and Campbell, 1980). Rats tend to solve this problem by using the distal cues in the environment to guide them to the different places on the maze. This deficit can also be seen in aversively-motivated tasks such as the Barnes circle plataform (Barnes, 1979) or the Morris water task (Morris, 1981). On the circular platform task rats must learn to go to one of 18 holes that surround the periphery of the brightly illuminated platform. A dark escape tunnel exists under this hole, and rats naturally prefer dark enclosed spaces to brightly lit open surfaces. They therefore learn in which direction from the central starting location the escape tunnel resides by learning its location in the testing room. Even though both age groups greatly improve their performance on this task over days, old animals are worse at remembering where the tunnel is from day to day than are the young animals (Barnes, 1979; Barnes et al., 1980). Furthermore, older animals forget this problem faster, after varying retention intervals, than do young animals (Barnes and McNaughton, 1985). The water task in its "spatial" configuration involves remembering the location of a hidden platform in a large pool of opaque water, again by using the distal cues available in the environment. Old animals can learn to find the platform within a few trials if the platform is above the surface of the water (Rapp et al., 1987). However, if they are forced to remember the submerged platform's location in the pool, they are worse at navigating to it than are young animals (Gage et al., 1984; Rapp et al., 1987).

Through a number of control procedures, it has been possible to conclude that the main contributor to the age deficit is likely to be problems with spatial memory rather than primarily sensory or motor factors. For example, old animals that have been well trained on this problem throughout their lives can navigate just as accurately on the radial maze as can young animals (Beatty, Bierley and Boyd, 1985; Bierley, Rixen, Troster and Beatty, 1986), suggesting that locomotor and visual factors are not limiting in this situation. Furthermore, certain drugs can improve the performance of old animals on the radial maze (Davis, Idowu and Gibson, 1983; de Toledo-Morrell, Morrell, Fleming and Cohen, 1984; Gallagher, Bostock and King, 1985), again suggesting that peripheral defects are not the sole contributors to the age deficit. In the water task the old rat's ability to swim to the platform if it is above the surface indicates that locomotor patterns involved in swimming are intact. Furthermore, if old rats are given an opportunity to solve a problem in a number of different ways, they will tend to solve the task using strategies that are *not* spatial. suggesting a strategy preference shift with age (Barnes et al., 1980; Rapp et al., 1987). This shift may be due to an inability of old animals to solve these kinds of problems effectively. In support of this conclusion is the finding that when old animals are trained on other versions of the radial maze problem that must be solved in a non-spatial manner, they are just as good as young animals in these tasks (Barnes, Green, Baldwin and Johnson, 1987).

Because damage to the hippocampal formation in young animals produces qualitatively similar spatial memory deficits (although quantitatively much more severe), the function of the hippocampus was measured electrophysiologically in awake unrestrained animals, or in the hippocampal slice preparation. Anesthetics were avoided in these age comparisons since they are known to differentially influence the age groups. It should be pointed out that the majority of biophysical properties of hippocampal cells are completely normal in old rats (Barnes and McNaughton, 1980a). However, there *are* a number of changes that will be emphasized here, primarily because they may underlie the cognitive deficits that we see in old animals.

The first difference noted in hippocampal plasticity between young and old rats was an alteration in their ability to maintain longterm synaptic enhancement (LTE). This kind of plasticity (e.g., Bliss and Gardner-Medwin, 1973; Bliss and Lomo, 1973; McNaughton, 1982; and see Teyler this symposium) can be induced in the hippocampus by brief bursts of high frequency stimulation of temporal lobe afferents, and results in the strengthening of synaptic communication in this pathway that can be measured using extracellular field potential recording techniques. Many neuroscientists believe that a mechanism similar to the one induced artificially by electrical stimulation is the one that normally operates to store information in the central nervous system. When a comparison of LTE was made between young and old animals, the absolute magnitude of the increase in synaptic strength was not different between age groups (Barnes, 1979). After the high frequency stimulation treatment the decay of LTE could be followed for weeks and was found to be faster in the older animals. That is, the decay time constant for LTE in the young animals was 37 days and for the old animals 17 days (Barnes and McNaughton, 1980b). Moreover, when the ability of an individual animal to perform on the circular platform task was compared to that animal's ability to maintain LTE at hippocampal synapses, a statistically significant correlation was found within age groups (Barnes, 1979). This, and a number of other lines of evidence, suggests that LTE may be the physical substrate of memory and that the mechanism for maintenance of this synaptic change may be defective in old animals. This may contribute to old animals' faster forgetting of spatial information (Barnes and McNaughton, 1985).

Another change between age groups that has been noted in the hippocampal formation is that there are approximately one-third fewer synaptic contacts from the entorhinal cortex onto hippocampal granule cells in old rats (Geinisman and Bondareff, 1976). This results in a smaller evoked synaptic field response recorded from granule cells in old rats (Barnes, 1979). This loss of synaptic input, however, does not result in a shutdown of information transfer through the hippocampal circuit of old rats. A lowering of the discharge threshold of the granule cells and an increase in the strength of the remaining synapses onto old granule cells nearly compensates or balances for the loss of synaptic input (Barnes and McNaughton, 1980a). Thus, the overall probability of output from the granule cells is preserved as a result of these changes. The exact mechanisms through which these changes are accomplished are not understood. However, we think that we may have a good explanation for the changes in cellular excitability. The most common mechanism for neuronal communication is via synaptic contacts involving the release of neurotransmitter substances. There is another important means by which cells can communicate, however, and that is via electrical junctions. One type of electrical junction, the gap junction, has been morphologically identified between hippocampal cells (MacVicar and Dudek, 1982; Schmalbruch and Jahnsen, 1982). Furthermore, these junctions can be examined through the use of low molecular weight fluorescent dyes. If the dye is injected into one cell, it will pass across any gap junction into connected cells (MacVicar and Dudek, 1980; Rao, Barnes and McNaughton, 1986). In this manner, it was shown that old hippocampal cells are more extensively interconnected via these gap junctions than are young cells (Barnes, Rao and McNaughton, 1987). The result of an increase in gap junctions would be that the electrically coupled cells would tend to fire with the cell in that group that had the lowest discharge threshold or that was most activated by the afferent input. This would result in an apparent lowering of the discharge threshold of the cell, and we think this may be at least a partial explanation for the excitability change that we observe in old hippocampus.

Although there may be compensatory mechanisms at work to keep the information processing characteristics of the hippocampus relatively normal, the amount of information coming into this structure from its main afferent input pathway is substantially reduced. It was predicted, therefore, that there should be a difference between young and old cells in terms of their information content. It has been known for some time that certain single cells can be recorded in the hippocampus that show their maximum firing rates only in very restricted areas of an environment (O'Keefe and Dostrovsky, 1971). Because they appear to fire in response to particular locations in a given space, they have been called "place cells" (O'Keefe, 1976). The prediction with respect to the activity of young and old place cells was that the old cells should be less specific and less reliable than the young cells, because they are firing on the basis of much less information or input. To assess this, young and old rats were trained to traverse a radial 8-arm maze for 8 trials (or 64 arm choices) per day. When a single unit was isolated, the rat was tested using this behavioral procedure which allowed for a good statistical sample of the cells' firing characteristics on this maze. A comparison of the firing properties of young and old rat hippocampal place cells revealed that, indeed, the information processing characteristics of the old cells were altered. The old cells fired over a greater portion of the maze surface and were not as consistent from trial to trial as were the young cells (Barnes, McNaughton and O'Keefe, 1983). This occurred in the absence of any change in the mean or peak firing rate characteristics or interspike-interval distributions of the cells between age groups. Therefore, the reduction in information coming into the hippocampus appears to have an important effect on the way in which these cells respond to environmental input.

The results from these kinds of electrophysiological and behavioral analyses suggest that spatial memory problems of old animals most likely result from a combination of changes in hippocampal neuronal properties. The first general property involves the old hippocampal cell's reduced ability to retain spatial information, and the second is a spatial representational deficit. The task for those of us using animal models of normal age-related memory declines is to more precisely relate these changes found in animals to those observed in normal human aging and to begin to develop methods to attenuate these deficits.

SUMMARY

Aged organisms show a decline in their ability to learn and remember in certain situations. For example, it appears that both aged humans and aged rats exhibit spatial memory deficits. It is more difficult for older organisms to learn about and to navigate accurately within a familiar environment than it is for younger organisms. Because the brain structure that is critically involved in this type of behavior (the hippocampus) is relatively well understood in the rat, a correlation of spatial behavior with hippocampal physiology has been particularly useful in the delineation of some of the potential brain changes responsible for memory changes with age. Evidence for an age-related deficit in spatial memory is presented that emphasizes the importance of the contribution of the spatial component to the learning/memory changes seen with age in rats. The contribution of changes in hippocampal synaptic plasticity and in the information processing characteristics of single hippocampal cells of old rats is also discussed in terms of the potential influence on old animals' performance on spatial tasks.

RESUMEN

Organismos envejecidos muestran un declinar en su habilidad para aprender y recordar en ciertas situaciones. Por ejemplo se muestra que tanto seres humanos envejecidos como ratas envejecidas muestran déficit en la memoria espacial. Es más difícil para seres mayores aprender a conducirse adecuadamente en torno a un ambiente familiar que lo que les sucede a los jóvenes. La estructura cerebral que está involucrada en este tipo de conducta (el hipocampo) es bien comprendido en la rata, una correlación de la conducta espacial con la fisiología hipocámpica ha sido particularmente útil en la declinación de algunos de los cambios potenciales del cerbero responsables de los cambios en la memoria con la edad. La evidencia de un déficit vinculado a la edad en la memoria espacial señala la importancia de la contribución del componente espacial a los cambios de aprendizaje y memoria observado con la edad en ratas. La contribución de cambios en la plasticidad sináptica hipocámpica y en las características del proceso de información de células hipocámpicas aisladas de ratas viejas es también señalado en relativo a la influencia potencial sobre animales viejos en los logros en las tareas espaciales.

RÉSUMÉ

Les organismes âgés font preuve d'un déclin de leurs possibilités à apprendre et à remémorer dans certaines situations. C'est ainsi par exemple, qu'aussi bien l'homme que le rat âgé ont un déficit de la mémoire spatiale. Pour les organismes âgés, il est plus difficile qu'aux plus jeunes d'apprendre à connaître un environnement et à s'y mouvoir avec précision. En raison du fait que la structure cérébrale qui est électivement impliquée dans ce type de comportement (l'hippocampe) est relativement bien connue chez le rat, une corrélation du comportement spatial avec la physiologie de l'hippocampe a été particuliérement utile dans l'identification de certaines des modifications des capacités cérébrales responsables des troubles de la mémoire du sujet âgé. La preuve d'un déficit de la mémoire spatiable en relation avec l'âge confirme l'importance de la composante spatiable dans les modifications de l'apprentissage et de la mémoire qui apparaissent chez le rat avec l'âge. La participation des modifications de la plasticité synaptique de l'hippocampe et des caractéristiques de l'information dans les cellules isolées de l'hippocampe de vieux rats est également discutée dans son influence potentielle sur les performances de l'utilisation de l'espace chez les animaux âgés.

ZUSAMMENFASSUNG

Alte Organismen weisen eine Verminderung der Lernfaehigkeit und des Gedaechtnises bei gewissen Zustaenden auf. Z.B. zeigt sich, dass sowohl aeltere Menschen als auch aeltere Ratten eine Verminderung der raeumlichen Erinnerujg aufweisen. Aelterden Wesen faellt es schwerer, sich angemessen im familliaeren Milieu zu verhalten las juengere. Die zerebralen Strukturen die bei dieser Art der Verhaltungsweise affektiert sind, ist der Hippocampus, ist bei der Ratte gut bekannt: eine Beziehung zwischen den raeumlichen Verhalten und der Physiologie des Hippocampus, ist besonders evident bei der Verminderung einiger Veraenderungen der Potentiale des Gehirns, die verantwortlich sind fuer die Gedaeechtnisverminderung mit zunehmendem Aler. Die Augdnscheinlichkeit eines Defizits in Verbindung mit dem zunehmenden Alter beim raeumlichen Gedaechtnis, zeigt die Wichtigkeit des Anteils der raeumlichen Komponente bei er Veraenderung der Lernfaehigkeit und des Gedaechtiniss was man bei Ratten mit zunehmenden Alter beobachtet hat Der Anteil der Veraenderungen der synaptischen Plastizitaet des Hippocampus im Charakte des Informationprozesses isolierter Hippocampuszellen von alten Ratten wird ebenfalls hervorgehoben bezueglich des potentiellen Einflusses, bei aelteren Tieren, auf die Erfolge bei raeumlichen Aufgaben.

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Mechanisms of Behaviorally-Elicited and Electrically-Elicited Long-Term Potentiation

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Introduction

This paper expands briefly upon two aspects of Dr. Teyler's review, both related quite specifically to research in my laboratory. The first has to do with behaviorally induced phenomena similar to high frequency activated longterm potentiation (LTP) and the second has to do with possible morphological synaptic substrates of LTP.

Persistence of Behaviorally Elicited LTP in the isolated Dentate Gyrus in vitro

Behaviorally-elicited LTP refers to a variety of studies in which the postsynaptic responses to afferent activation is altered as a result of training or some other behavioral experience (e.g. Sharp et al., 1985). The issue of whether behaviorally-elicited LTP is a manifestation of the same process as is evoked by high frequency afferent activation (e.g. Bliss and Garner-Medwin, 1973; Bliss and Lmo, 1973; Racine et al., 1983) remains unresolved. However, evidence is converging to support this view. One aspect of electrically-elicited LTP is that it can be elicited and maintained in the hippocampal slice *in vitro.* This indicates that electrically-elicited LTP in the hippocampal formation need not depend, for either its elicitation or its maintenance, upon any region of the brain outside the hippocampal formation.

Behaviorally-elicited LTP certainly must depend for its elicitation upon input from other brain regions, especially the perforant pathway from the entorhinal cortex, which shows potentiation. However, the issue of whether behaviorally-elicited LTP remains in the absence of intact afferent systems is an important aspect of the possible commonality of features, and by implication, underlying mechanisms, of behaviorally-elicited LTP and electrically-elicited LTP. This issue is not a trivial one, since the hippocampal formation is subject to a number of phasic and tonic influences from extra-hippocampal afferent systems (e.g. Alvarez-Leefmans and Garner-Medwin, 1975; Winson, 1980, 1981).

A second aspect of the comparability of behaviorally-elicited and electrically-elicited LTP involves the underlying mechanism. Electrically-elicited LTP in the dentate gyrus involves a change in the efficacy or the number of synapses between axons of the perforant path, which receives the stimulation, and dendrites of the dentate gyrus granule cells, the responsiveness of which is altered by stimulation. It is important to determine whether the change

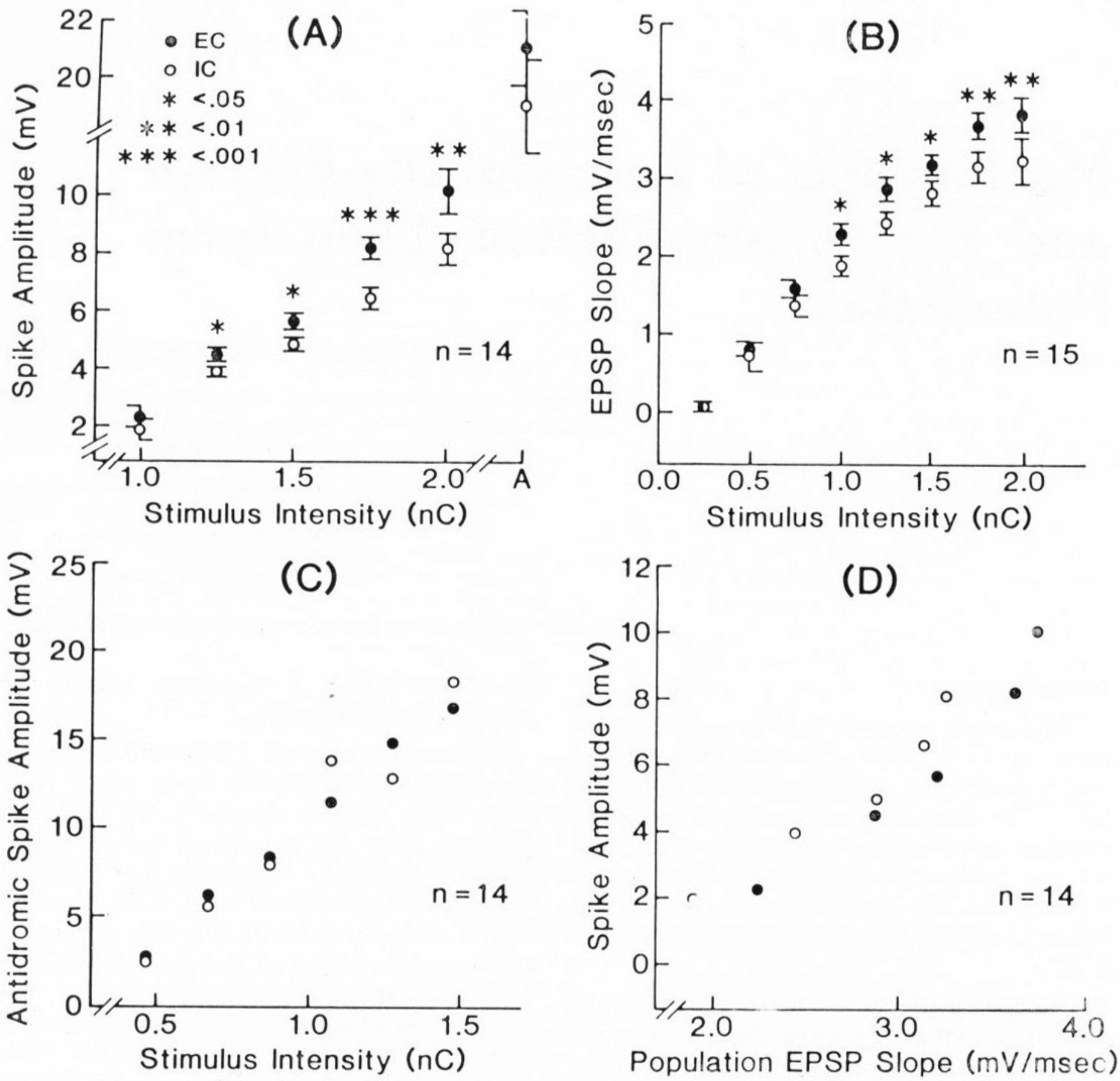

*Fig. 1. — Immediate effects of rearing condition on medial perforant path synaptic transmission (granule cell layer recordings). Stimulus intensity is expressed as the product of the stimulus current and duration; "A" denotes asymptotic value. The standard errors of the means are illustrated. * p < .05; ** p < .01; *** p < .001. A. Population spike input-output relations. B. Population EPSP input-output relations. C. Input-output relations for the antidromic population spike. D. Population spike/EPSP relations. (From Green and Greenough, 1986; c. 1986, The American Physiological Society).*

underlying behaviorally-elicited LTP is at the same location and shares details of physiological characteristics.

These issues have been investigated by Green and Greenough (1986). Rats were reared from weaning for approximately 30 days in either individual plastic tub cages (IC) or environmental complexity (EC), which consisted of a group of 12 rats housed in a large mesh cage filled with a set of "toys" (various wood, metal, and plastic children's toys and similar items) which was changed each day. EC rats also had the opportunity to explore a large "playbox" filled with another novel set of toys for 30-60 minutes each day. The hippocampus was removed from these animals and slices were studied *in vitro*. The population excitatory postsynaptic potential (EPSP) and the population spike (PS) were recorded in the granule cell layer of the dentate gyrus in response to stimulation of the medial perforant path. Both PS amplitude and EPSP slope were greater in EC rats across a range of stimulus intensities (see Figure 1). There was no significant difference between groups in the response to antidromic stimulation and no difference in the size of the presynaptic fiber volley recorded in the molecular layer afferent region, indicating that the altered responsiveness was due to altered efficacy or number of perforant path-granule cell synapses, as is also true for electrically-elicited LTP. A second experiment indicated that the difference between groups decayed to non-detectable levels when the EC rats were rehoused for an additional 3-4 weeks in IC cages. Thus the results indicated that both in terms of confinement to the isolated hippocampal formation and in terms of apparent synaptic mechanism, behaviorally-elicited LTP appears similar to electrically-elicited LTP in the dentate gyrus. Moreover, the time course of decay of the two phenomena must be at least roughly comparable.

Synaptic Morphological Correlates of LTP

There are a number of hypotheses as to the mechanisms underlying electrically-induced (and, ideally, also behaviorally-induced) LTP. Since there are at least 2 components to electrically-induced LTP, one lasting minutes to hours, the other lasting over days in the hippocampal formation (Racine et al., 1983), there is room for more than one mechanism. Perhaps the more interesting form of LTP, at least in terms of its relevance to long term memory, is the longer-lasting component. Many of the hypotheses, including changes in the number of synapses as well as several proposals for changes in the efficacy of previously-existing synapses, make morphological predictions. Evidence for several different forms of morphological change has been presented in different hippocampal formation systems. Here, continuing the parallel between behavioral and electrical elicitation of the phenomena, the types of synaptic morphological changes in the CNS in general arising from behavioral stimulation and from electrical elicitation of LTP in the hippocampal formation are compared.

The first proposed morphological basis for efficacy change associated with LTP was spine shape change. Van Harreveld and Fifkova (1975) reported increased spine area and perimeter in dentate gyrus synapses in response to high frequency perforant path activation. This result is compatible with the proposal that the efficacy of synaptic transmission could vary with the resistance of the postsynaptic spine, where the size of the stem would be a critical component (Rall, 1970, 1978; Wilson, 1984). Subsequent reports indicated that this swelling persisted for at least 23 hours following the LTP induction procedure, confirmed the induction of LTP and indicated that spine stems, the high resistance region in the Rall model, are both shortened and widened as a consequences of LTP induction (Fifkova and Van Harreveld, 1977; Fifkova and Anderson, 1981; Fifkova *et al.*, 1982; Payne *et al.*, 1982). Desmond and Levy (1983) similarly reported an increase in the relative frequency of large post-synaptically concave spines in the region of maximal synaptic activation. These findings were interpreted as indicating spine swelling, supporting the Fifkova work.

In hippocampal subfield CA1, LTP stimulation via Schaffer collaterals produced no differences in the mean area of dendritic spine profiles or in the widths of spine stems, although there was less variation in these measures in potentiated preparations (Lee *et al.*, 1980; Lee *et al.*, 1981). Chang and Greenough (1984) repeated these observations and demonstrated, using the stereological ratio of perimeter to area, that spine heads became rounder following LTP in the *in vitro* CA1 preparation, and that there were no differences in spine profile area, in headed spines. The shape change had disappeared by 8 hours after LTP stimulation, whereas significant LTP, measured electrophysiologically, remained. Moreover, the shape change was not evident statistically when quick freezing tissue preparation was used (Chang et al., 1986). Thus the shape change does not account entirely for LTP in the Schaffer collateral system, although whatever results in altered shape in aldehyde preparations may be involved in shorter term aspects of long term potentiation.

The dentate gyrus and area CA1 may differ in utilization of spine shape change as a plasticity mechanism. Certainly there is variation in the evidence that spine shape change occurs in response to behavioral situations. Coss and Globus (1978) reported that spine density was greater and spine stems were shorter on ascending dendrites of optic tectum interneurons in socially maintained *vs.* isolated jewel fish. A similar shortening of spine stems reportedly occurs on corpora pedunculata interneurons during the progression from nurser to forager in honeybees (Coss et al., 1980). Brandon and Coss (1982) have reported that detectable spine stem shortening occurs as a consequence of a few minutes of "orientation flight" in the honeybee (see also Perkel and Coss, 1984). In contrast to these supportive results, Sirevaag and Greenough (1985) used electron microscopy to measure length, perimeter, area, and roundness of heads of spines in visual cortex of rats reared in complex, social, and individual environments and found no statistically significant group differences.

There is evidence for a presynaptic component of LTP efficacy change (Skrede and Malthe-Sorensen, 1981; Dolphin *et al.*, 1982; Anderson *et al.*, 1984). Some recent evidence indicates an increase in the number of vesicles near the presynaptic membrane in electrically potentiated hippocampal preparations (Applegate *et al.*, 1987; Desmond and Levy, 1986). This evidence is exciting because it may correspond to changes associated with increased synaptic efficacy in invertebrate preparations (Bailey and Chen, 1983). However, in another model system, there is no indication of differential numbers of equivalently-positioned vesicles in neuromuscular junctions differing in synaptic efficacy (Herrera *et al.*, 1985). Moreover, the interpretation of these results is clouded not only by the controversy regarding the role of vesicles in synaptic transmission (e. g., Tauc, 1982; Oorschot and Jones, 1987) but also by the fact that the aldehyde fixatives used in these studies depolarize neuronal processes and cause neurotransmitter release (McKinlay and Usherwood, 1978), such that confirmation by quick-freeze procedures such as those used in the original Van Harreveld and Fifkova (1975) study would be valuable. In addition, in contrast to these reports, Schuster *et al.* (1986) reported a decrease in vesicle density near the presynaptic membrane following LTP treatment in the dentate gyrus. In what could be a related phenomenon, there are several reports that both visual deprivation and environmental complexity manipulations can affect vesicle measures (Vrensen and DeGroot, 1974, 1975; Sirevaag and Greenough, in press) with similar measures apparently affected in LTP studies (Applegate *et al.*, 1987), such that comparison of aldehyde and quick-freeze data seems worth the effort.

The morphological LTP correlate that shares the greatest commonality with behaviorally-induced morphological change is the formation of synapses. Both Lee et al. (1980, 1981) and Chang and Greenough (1984, 1986) have reported increased synaptic density, indicating the formation of new synapses, in subfield CA1 of the hippocampus following the induction of LTP. Chang and Greenough reported that synapse formation occurs within 10-15 minutes or less, a time compatible with the temporal parameters of LTP. Synapse formation or retention (or related measurement of dendritic extent) is similarly affected by behavioral manipulations such as dark-rearing (e. g. Cragg, 1975), juvenile or adult exposure to a complex environment (Turner and Greenough, 1985; Beaulieu and Colonnier, 1985; Hwang and Greenough, 1986), and behavioral training (Chang and Greenough, 1982; Greenough et al., 1985a). There is also evidence that the formation of synapses is involved in long-term forms of plastic change in invertebrates (Bailey and Chen, 1986). Other morphological evidence has been interpreted as indicating that synapses may form in response to behaviorally-induced neural activity (Greenough et al., 1985b). Synapse formation is also compatible with other physiological changes reported to be associated with LTP induction, such as increased neurotransmitter release (Skrede and Malthe-Sorensen, 1981; Dolphin *et al.*, 1982) and increased receptor numbers (Lynch and Baudry, 1984). Thus it remains perhaps the most likely basis for LTP and one of the most likely candidates for involvement in long-term memory in general.

Acknowledgements

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SUMMARY

Electrically-elicited LTP in the hippocampal formation need not depend for either elicitation or its maintenance, upon any region of the brain outside the hippocampal formation. Behaviorally-elicited LTP certainly must depend for its elicitation upon input from other brain regions, specially the perforant pathway from the entorhinal cortex showa potentiation.

Certainly there is variation in the evidence that spine shape change occurs in response to behavioral situations.

Lee et al and Chang and Greenough have reported incresed synaptic density, indicating the formation of new synapsee in subfield CA1 of the hippocampus following the induction of LTP. Synapses may form in responses to behaviorally-induced neural activity.

Synapse formation is also compatible with other physiological changes reported to be associated with LTP induction such as increased neurotransmitter release and increased receptor numbers. Thus it remain perhaps the most likely basis for LTP and one of the most likely candidates for involvement in long-ferm memory in general.

RESUMEN

LTP promovido electricamente en la formación hipocampica no está en dependencia para su promoción o su mantenimiento, de ninguna región del cerebro fuera de la formación hipocámpica. LTP inducido por conducta depende para su promoción de influjos de otras regiones del cerebro, especialmente la vía perforante de la corteza entorinal, la cual muestra potenciación.

Ciertamente existe una variación en la evidencia que el cambio en la forma de la espina ocurre como respuesta a situaciones de conducta.

Lee y otros y Chang y Greenough han reportado densidad Sináptica acentuada indicando la formación de nueva sinapsis en el subsuelo CA1 del hipocampo siguiendo la inducción de LTP. Las sinapsis pueden formarse en respuesta a la inducción de actividad neural por conducta. La formación sináptica es también compatible con otros cambios fisiológicos asociados con inducción de LTP tales como acentuación de la Liberación de neurotransmisores y aumento del número de receptores. De este modo queda quizá la base más probable para LTP y uno de los probables factores que están involucrados en general en la memoria a largo plazo.

RÉSUMÉ

La LTP provoqué électriquement dans l'hypocampe n'est sous la dépendance d'aucune région cérébrale autre que le systéme de l'hypocampe.

La LTP induite par conduction dépand pour sa propagation de l'influence d'autres régions cérébrales, et particuliérement la voie perforente du cortese "entorhinal" potentialisé.

Tres certainement il existe une variation de forme de la "spicule", comme réponse a une situation de conduite.

Lee et col., Chang et Greenough ont rapporté des dansités synaptiques augmentées,

ce qui indique la formation de nouvelles synapses dans le CAL de l'hypocampe, et qui suivent les inductions de LTP.

Les synapses peuvent se former comme réponse a l'induction de l'activité neurale par conduite.

La formation synaptique est compatible avec d'antres changements phisiologiques associés avec l'induction LTP, tels que l'augmentation de la liberation de neurotransmieteurs et l'augmentation de la quantité de récepteurs. Tell est peut être la base probable du LTP et un des facteurs en relation en général avec la memoire a long terme.

ZUSAMMENFASSUNG

Elektronisch potenzierter LTP im Hippocampus braucht nicht a bzuhaengen von irgend einer ausserhalb des H. liegenden Gehirnregion, weder zu seiner Produktion noch zu seiner Aufrechterhaltung. Die durch die Verhaltungsweise produzierte LTP haengt bezueglich ihrer Produktion von den Linflussen anderer Regionen des Gehirns ab, besonders die via perforanz, der entorhinale Cortex weist Potenziation auf.

Gewiss besteht eine Variation in der Augenscheinlicheit, dass die Form des Dornes mit der Reaktion auf Verhaltenssituationen wechselt. Lee u. Mitarb. u. Chang u. Greenough haben berichtet ueber starke Dichte der Synapsis, was auf die Neubildung von Synapsis auf dem Boden des CAL des Hippocampus nach Induktion der LTP hinweist. Die Synapsis koennen sich bilden als Reaktion auf die neurale Aktivitaet durch die Verhaltung weisen. Die synapsi sbildung stimmt auch ueberei mit anderen physiologischen Veraenderungen durch die Induktion der LTP, wie die Vermehrung der Freisetzung von Neurotransmittern u. der Verhrnehrung der Anzahl der Rezeptoren. Auf diese Weise besteht vielleicht die Grundlage der LTP u. einer der moeglichen Faktoren, die im allgemeinen das Langzeitgedaechtinis betreffen.

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Angiotensin Converting Enzime and Memory: Preclinical and Clinical Data

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INTRODUCTION

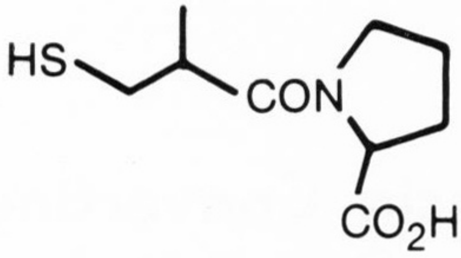
This report focuses on data from preclinical and clinical studies using captopril and other derivatives of the aminoacid proline (see Figure 1) that inhibit the angiotensin converting enzyme (ACE).

The ACE is a metalloexopeptidase carboxyhydrolase (EC 3.4.15.1) that catalyzes cleavage of dipeptides from the C-terminal end of peptide substrates (Soffer, 1976). Since its presence in homogenates of rat and human brain was observed (Yang and Neff, 1972; Poth et al 1975), it has been found in many areas of the brain. Relatively high levels of activity have been reported in the striatum, cerebellum, and pituitary gland (Yang and Neff, 1972; Benuck and Marks, 1979), the caudate and nucleus habenularis medialis (Poth et al, 1975; Saavedra et al, 1982), the area postrema, substantia nigra, locus ceruleus, globus pallidus and hippocampus (Chevillard and Saavedra, 1982; Defendini et al, 1983). In addition, ACE is highly concentrated in the choroid plexus (Erdos, 1985; Igic et al, 1977; Defendini et al 1983) and has been demonstrated in cortical brain microvessels (Gimbrone et al, 1979; Brecher et al, 1981) as well as in the cerebrospinal fluid (Schelling et al, 1980; Kariya et al, 1982; Lantz and Terenius, 1985).

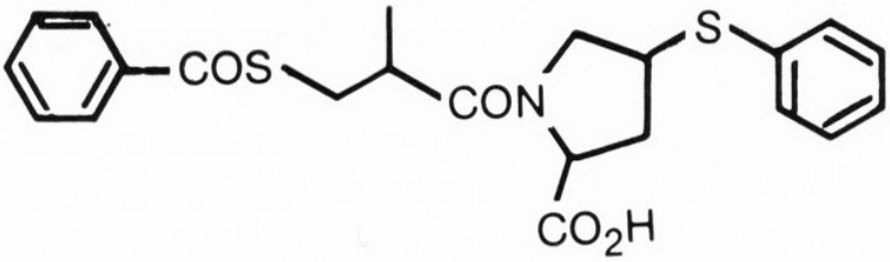
Recent studies provide convincing evidence that ACE is relatively selectively found in neuronal cells (Lentzen et al, 1983; Palenker et al, 1984; Koshiya et al, 1984, 1985; Mizuno et al, 1985) and that it is located on terminal membranes (Velletri and Lovenberg, 1983) which suggest its involvement in synaptic transmission. Moreover, the relevance of its discrete localization in the brain as well as charges in its activity has been raised in regard to schizophrenia (Arregui et al, 1979; Beckman et al, 1984) and other pathological conditions such as Huntington's chorea (Arregui et al, 1977, 1978), progressive supranuclear palsy, Parkinson's disease, and Alzheimer's disease (Arregui et al, 1982; Zubenko et al, 1985, 1986) which share the ability of affecting cognitive memory processes.

Substrate specificity studies have shown that ACE primarily catalyzes the formation of the potent endogenous vasoconstrictor octapeptide angiotensin II (AII) from the relatively inactive decapeptide angiotensin I and that it is involved also in the metabolic degradation of bradykinin, enkephalins, substance P, and neurotensin (Cushman et al, 1981; Prinz et al, 1982; Marks et al, 1980; Lee et al, 1979; Checler et al, 1983). It is possible, therefore, that at least partially the antihypertensive action of the

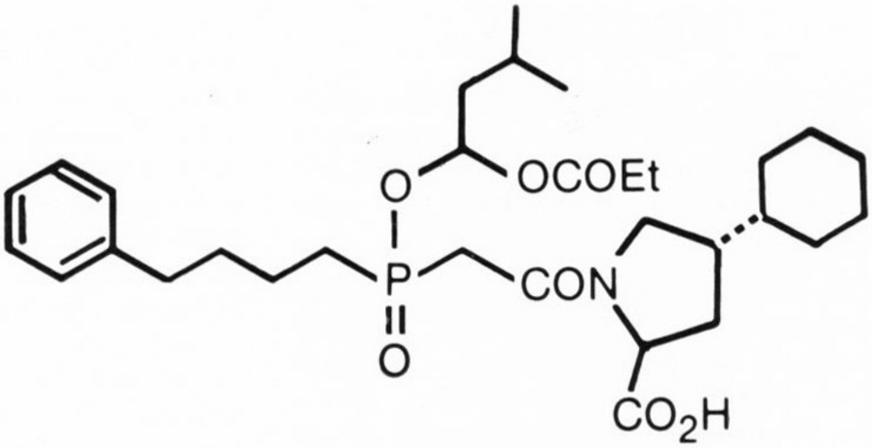
Captopril (SQ 14,225)



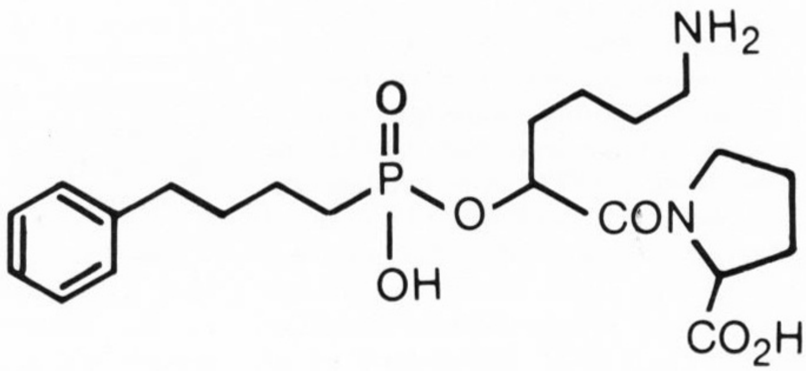
Zofenopril (SQ 26,991)



Fosinopril (SQ 28,555)



SQ 29,852



inhibitors of ACE may be due to accumulation of bradykinin after inhibition of kininase II which is identical with ACE (Erdos, 1975), and that alteration of various peptide/protein systems (including opiod peptides, vasopressin, substance P, enkephalins) may play a role in the mediation of the effects of ACE inhibition in different target organs, one of which is the brain.

The rather broad substrate specificity of the ACE, the fact that AII can elicit behavioral effects including disruption of passive avoidance behavior and inhibition of retention performance (Prinz et al, 1982), and the finding of increased levels of ACE activity in brains of Alzheimer's disease patients (Arregui et al, 1982) provided grounds for exploring the involvement of the enzyme in cognitive functioning.

PRECLINICAL STUDIES

Initially we examined the effects of captopril versus saline in learning and memory paradigms including habituation of the rearing response, light-dark discrimination and reversal learning in the rat, and as compared to saline and methyldopa in the delayed response performance and conditioned avoidance behavior models. While methyldopa significantly impaired performance in the delayed response paradigm, captopril was devoid of negative effects in this and in the habituation of the rearing response, the light-dark discrimination and the reversal learning models (Sudilovsky and Turnbull, 1982, unpublished). Findings with the conditioned avoidance behavior model were of particular interest in relation to ACE and are described in some detail below.

All animals used in these and the following series of studies were male Sprague-Dawley rats (Charles River, Wilmington, MA) 25 weeks old and weighing 300-450g. They were housed separately in stainless steel cages with continuous access to food and water on a 12 hour light/dark cycle (light: 1800-0600 hr.) at a constant room temperature (22° - 24° C). *All training and* testing was performed approximately 6 hours into the dark component of the light/dark cycle in a dimly lighted soundproof room using a standard shuttle box device (Lehigh Valley Electronics #146-04), a plexiglass chamber (50 x 20 x 20 cm) divided by a center barrier 7 cm in height. The conditioned stimulus consisted of a 10-second tone provided by a Sonlert mounted in the midpoint of the ceiling of the chamber. Floor current of 0.8 mA was delivered by a constant current shocker-scrambler (Lehigh Valley Electronics #133-3). The day before the initial trial each animal was allowed to explore the experimental chamber for 10 minutes without tone or shock. Injections were all administered intraperitoneally (i.p.) in a volume of 1 mg/ml and in blind fashion.

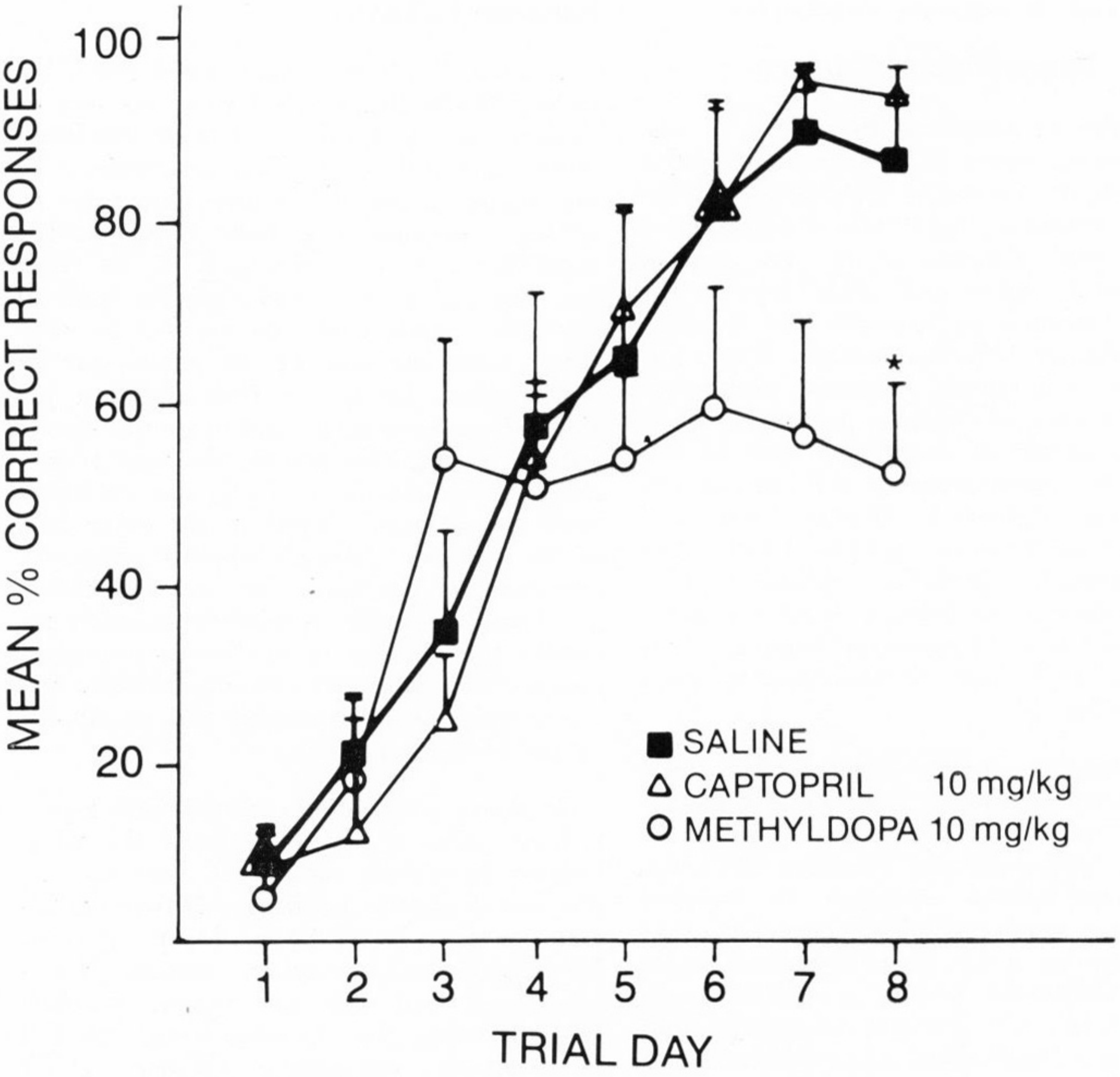
Acquisition of Conditioned Avoidance Behavior (CAA)

A total of 19 rats were tested for CAA during the eight days following the day of exploration. Animals received randomly either captopril 10 mg/kg, methyldopa 10 mg/kg, or saline on the two days prior to testing and then one hour before it for eight days. Testing consisted of 20 trials per day on a 30-second variable interval schedule. Shock could be avoided by shuttling from one side of the center barrier to the other during the 10-second tone period. If an animal did not cross the center barrier during this period, the tone remained on and the floor shock was delivered until the animal escaped to the other half of the chamber. Animals which consistently remained on the center barrier were removed from the study. Automatic counters recorded the number of avoidance responses, escapes, and intertrial crossings while a running time meter recorded the shock duration for each animal.

Two-way analysis of variance with repeated measures of the data from the trials yielded an overall significant difference in the rate of shuttle acquisition between treatment groups, $F(2,128)=3.64$; $p=0.0290$. No significant interaction between treatment and trial day was found. Post-hoc *analysis using the Newman-Keuls method* for multiple comparisons (Winer, 1971) demonstrated a significant difference between both the saline and methyldopa ($p<0.05$) and the captopril and methyldopa groups ($p<0.02$). No difference was found between the saline and captopril groups (see Figure 2).

Therefore, captopril had no effect on CAA while methyldopa impaired it significantly as compared to both saline and captopril. The possibility of such impairment resulting from a deficit in motor activity, coordination or motivation was highly unlikely since methyldopa did not affect intertrial crossings or escape latency.

ACQUISITION OF CONDITIONED AVOIDANCE BEHAVIOR



* $p < 0.05$ versus saline; $p < 0.02$ versus captopril

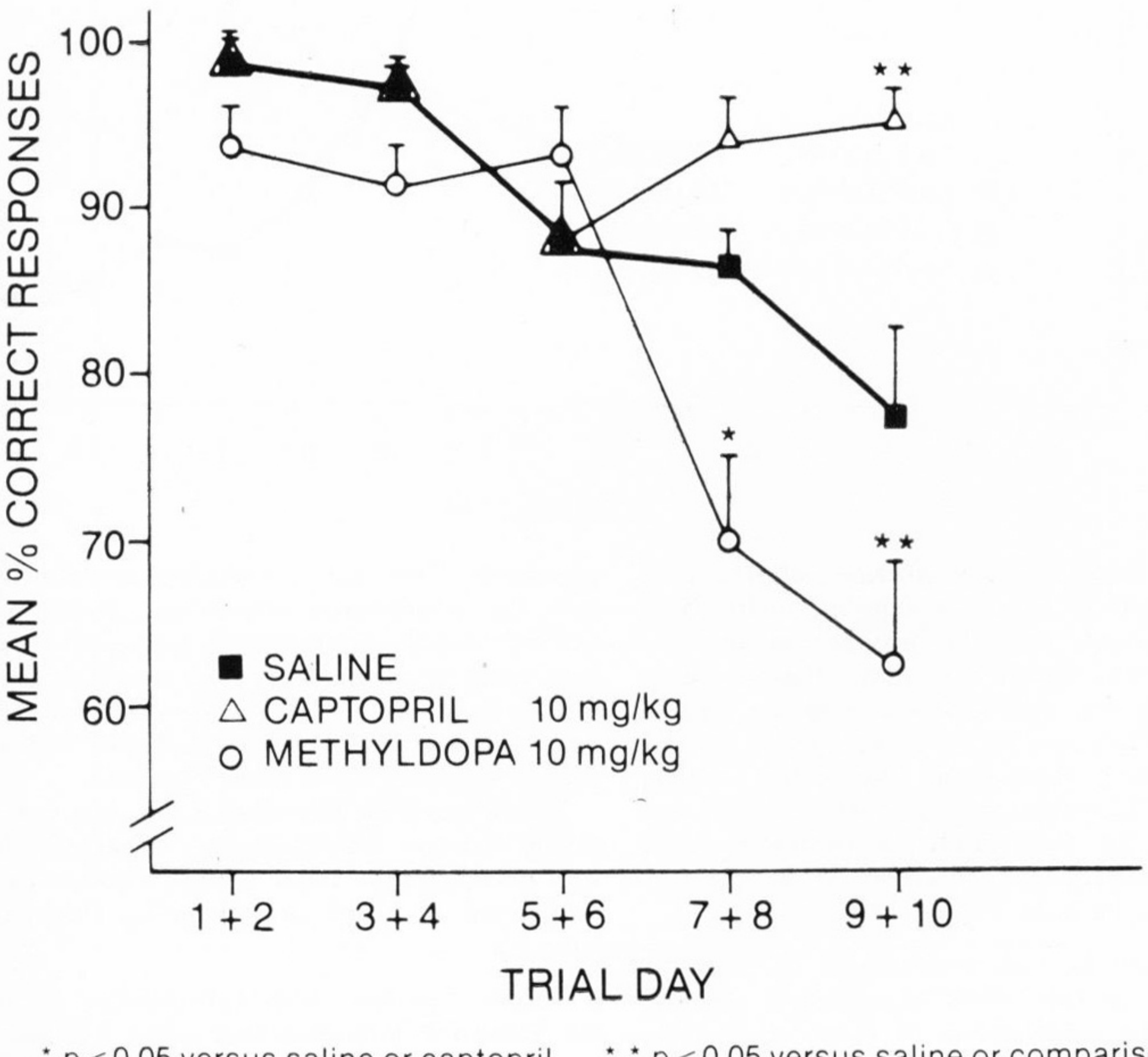
Extinction of Conditioned Avoidance Behavior (CAE)

In the second series of experiments animals were trained for the 15 days following the day of experimental chamber exploration. Each animal received 20 trials per day on a 30-second variable interval schedule as described above. No drug treatment was administered during the training period. Animals not meeting the admittance criterion of correct avoidance responding on at least 85 % of the trials for four out of the last five days of training were removed from the study. A total of 18 rats reaching the admittance criterion were tested for CAE during 10 days and received their randomly assigned treatment (captopril 10 mg/kg; methyldopa 10 mg/kg; or saline) on the two days prior to testing and then one hour before it. Testing consisted of 20 trials per day identical to those previously described, except that no shock was administered if an animal failed to shuttle during the 10-second tone period. The tone was simply discontinued and the testing proceeded.

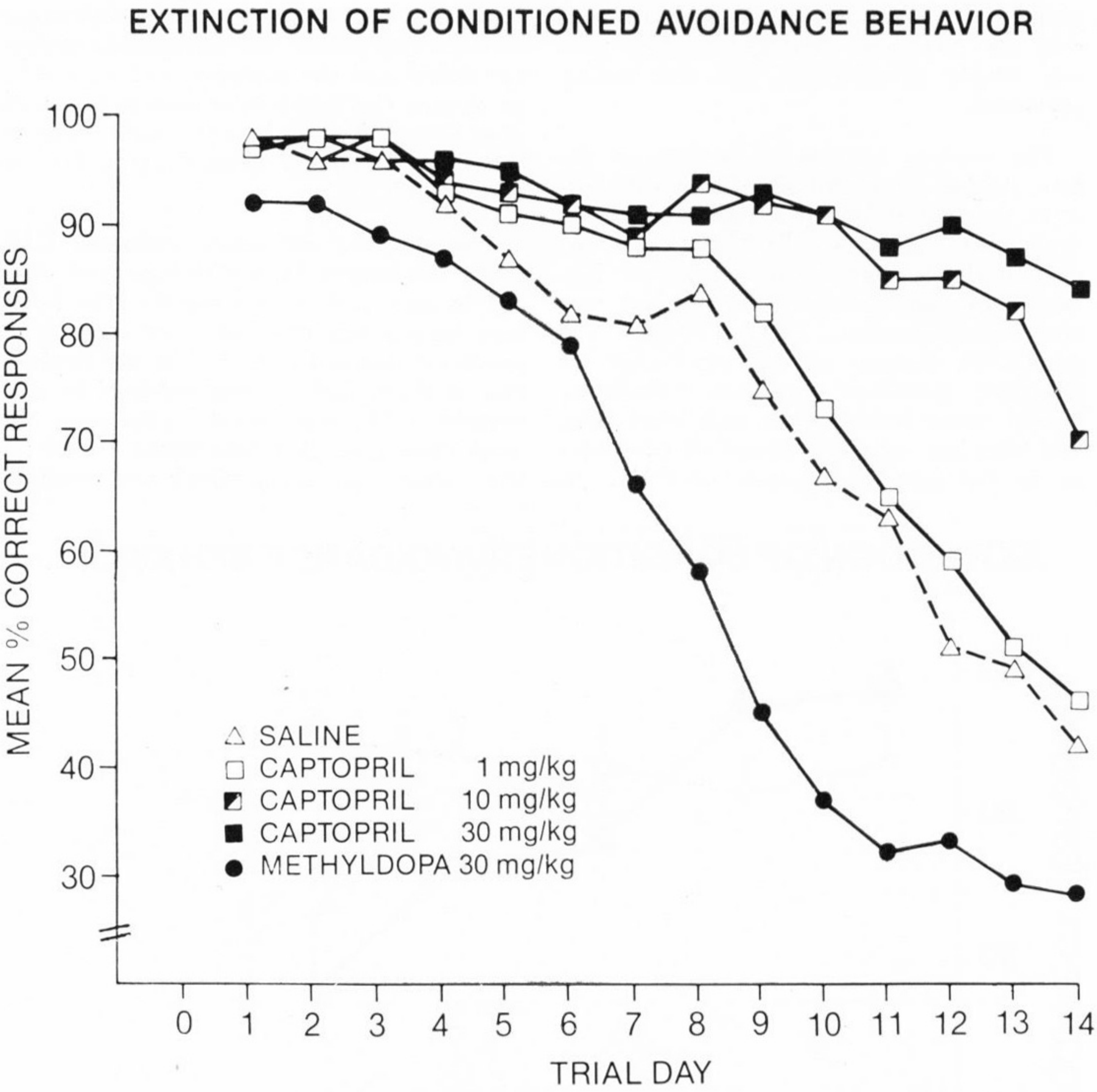
The two-way analysis of variance of the data yielded an overall significant difference in the rate of shuttle extinction between treatment groups, $F(2,150) = 9.56$, $p = 0.0001$. A significant interaction between treatment groups and trial days was also demonstrated, $F(18,150) = 1.75$, $p = 0.0368$. Because of this significant interaction, post-hoc analyses (Newman-Keuls) were restricted to individual days and were not collapsed across all trial days as in the post-hoc analysis of CAA. As shown in Figure 3, significant differences between the saline and methyldopa groups ($p < 0.05$) and the captopril and methyldopa groups ($p < 0.05$) were obtained on trial days 7 and 8, and between each other of the three treatment groups ($p < 0.05$) on days 9 and 10.

The observed differences indicated CAE to be accelerated by methyldopa and delayed by captopril, suggesting that the latter may have a protective effect on memory of previously learned tasks in the rat Replication of these findings was obtained in subsequent CAE experiments employing the same procedures described above except for the duration of testing which was extended

EXTINCTION OF CONDITIONED AVOIDANCE BEHAVIOR



* $p < 0.05$ versus saline or captopril   ** $p < 0.05$ versus saline or comparison drug



to 14 days, the i.p. dosages administered (captopril 1, 10, or 30 mg/kg; methyldopa 30 mg/kg), and the number of animals per group, which was nine. Post-hoc analyses of the data demonstrated no significant differences between captopril 1 mg/kg and saline throughout the entire testing period. However, captopril at both 10 and 30 mg/kg significantly protected CAE while methyldopa hindered it on days 9 through 14 (see Figure 4).

Support for the involvement of ACE inhibition in this effect of captopril was obtained by examination of three other inhibitors of the ACE in the same series of experiments. The captopril analog, zofenopril, and the phosphorous-containing fosinopril and SQ 29,852 administered i.p. on the two days prior to testing and then one hour before it at doses of 10 mg/kg each for 14 days were as active as 10 mg/kg of captopril in delaying CAE (see Figure5).

Epicaptopril on the other hand, the practically inactive diastereoisomer of captopril, was not different from saline whether administered at 1, 10 or 30 mg/kg (see Figure 6).

Taken together with information from the literature indicating that brain AII may be responsible for disruption of passive avoidance behavior in rats (Koller et al, 1979), and that when administered into the brain the angiotensin peptides have disruptive effects on retention performance in rodents (Melo and Graeff, 1975; Morgan and Routtenberg, 1977), the above results provide evidence to suggest that as well as other components of the renin-angiotensin system the ACE is involved in the physiological processes underlying learning and memory.

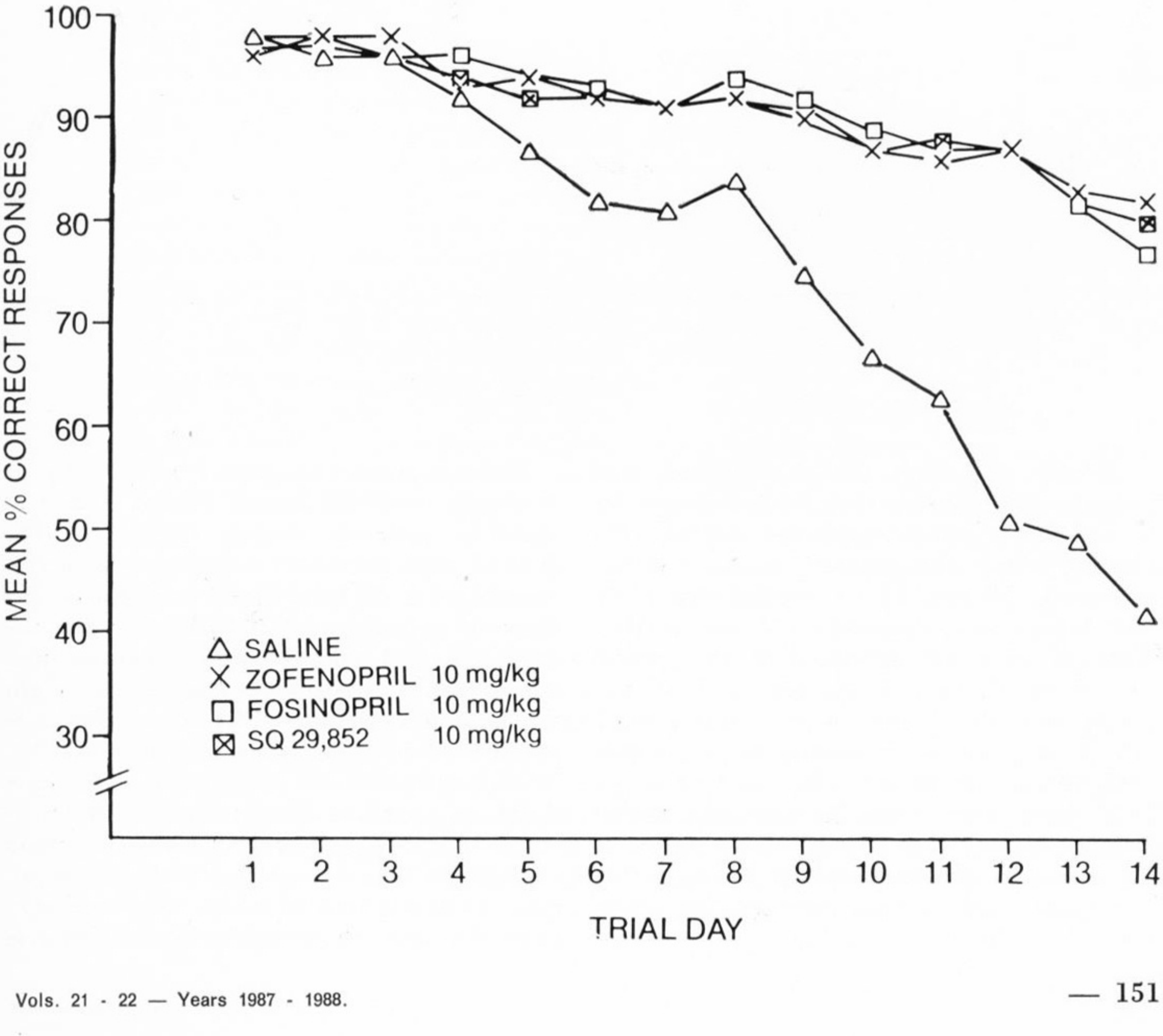
CLINICAL STUDIES

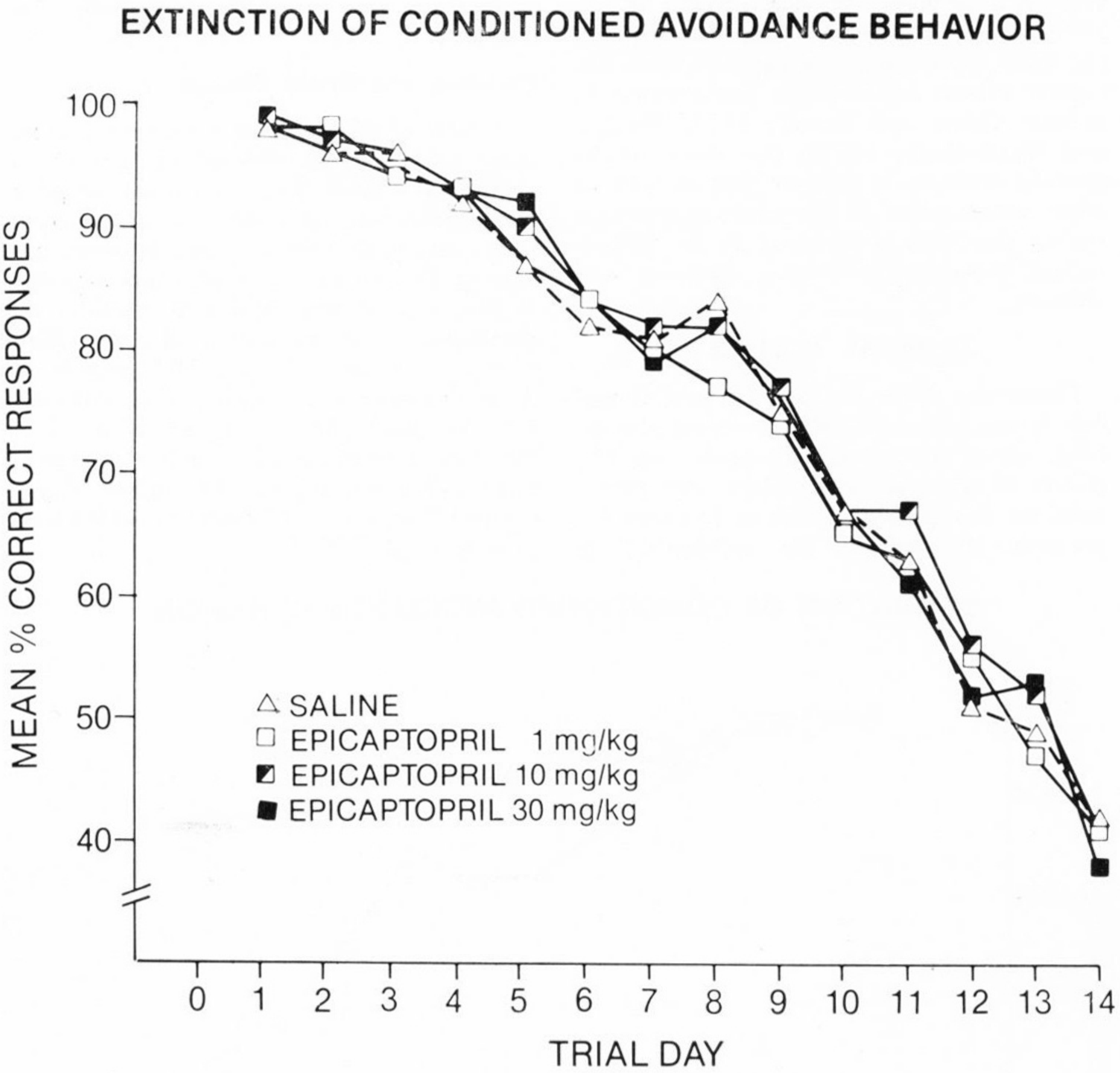
Given the above findings in animal models it was of interest that a recent clinical trial, aimed primarily at assessing the effects of captopril, methyldopa, and propanolol on the quality of life of hypertensive patients, afforded us the opportunity of evaluating also cognitive performance in humans.

Patients and Study Design

A total of 761 patients were recruited for these trials by 30 clinical centers throughout the United States. In order to generate a relatively homogeneous sample, only fully employed white males between the ages of 21 and 65 years who had a primary diagnosis of uncomplicated essential hypertension (median seated diastolic blood pressure between 92 and 109 mmHg on three determinations within five minutes) were enrolled. The methods employed in the clinical trial, including patient inclusion criteria and a complete description of procedures have been reported elsewhere (Croog et al, 1986).

EXTINCTION OF CONDITIONED AVOIDANCE BEHAVIOR





Briefly, the study design consisted of a one-month placebo period (A), followed by a six-month active treatment period (B) during which the patients received either captopril (50 mg. b.i.d.), methyldopa (500 mg, b.i.d.), or propranolol (80 mg, b.i.d.). Patients were not informed of the timing or length of the placebo period. Both pa tients and physicians were unaware of which drug was given during the six-month active treatment period. The standard step-2 level doses were chosen to maximize therapeutic efficacy with a single drug program. To maintain the double-blind during active treatment, each patient received the assigned drug plus two placebos.

Patients were evaluated medically at two-week intervals during Period A and at monthly intervals during Period B. The goal of antihypertensive therapy was achievement of a diastolic blood pressure of less than 90 mmHg. In accordance with the study protocol, patients whose diastolic pressure was 95 mmHg or greater at eight weeks into Period B (B8), or at any subsequent visit, were also administered hydrochlorothiazide 25 mg, b.i.d. This dose could be varied or discontinued by the physician at subsequent visits, depending upon changes in blood pressure. Patients requiring hydrochlorothiazide were excluded from the analyses of cognitive data because of uncertainties regarding the effects of both unregulated blood pressure and the direct or interactive effects of diuretic medication on cognitive processes.

Cognitive Measures

Because of the broad range of quality of life outcome measures (Croog et al, 1986) only a brief cognitive test battery could be employed. The measures chosen for this battery were the Visual Reproduction Test (VRT) from the Wechsler Memory Scale (Wechsler, 1945) and the Trail Making Test from the Halstead-Reitan test battery (Reitan, 1958). These are standard neuropsychological tests which have been used in previous studies of drug effects (Elias and Streeten, 1980), and which were chosen because of their sensitivity to modest, generalized organic impairments such as those that may be associated with antihypertensive therapy (Lezak, 1983). The Visual Reproduction Test requires patients to draw geometric designs from memory after these are shown for ten seconds and then removed from view. The Trail Making Test consists of two tasks. In the first task, part A (TMA), patients are required to connect in numerical order 25 numbers printed and distributed randomly on a blank page. The second part of the test, part B (TMB), requires patients to alternate between consecutive numbers and consecutive letters, i.e., they must connect the digit "1" to the letter "A" to the digit "2" to the letter "B", and so on. The time required to complete the task is the measure of interest for both the TMA and TMB.

Because both tasks involve sustained visuospatial scanning ability and directed visual shifting, they require visual attention and sustained concentration for successful performance. In general, TMA is considered to reflect psychomotor speed while TMB taps more complex cognitive abilities, especially those involved in rapidly shifting from one concept to another (Lezak, 1983). In addition, an index may be constructed by subtracting scores on TMA from those on TMB to form a measure of cognitive processing speed that is largely independent of psychomotor speed. Normative data for different age groups on TMA and TMB are available (Davies, 1968; Harley et al, 1980).

All tests were administered at the end of the placebo period and after 8 and 24 weeks of active treatment (B8 and B24). Registered nurses or other technical or administrative health personnel administered the tests. Testers were not directly involved with the care of the participating patients and were blind to study medication. Standardized training in psychological testing was given to all testers.

Statistical Analyses

Analyses of the data was focused on differences among the three drug groups from baseline to visits B8 and B24, and from visit B8 to visit B24 Patients with incomplete cognitive data were excluded from the analyses. Initially comparison was made of change in scores over time and by repeated measures analysis of variance with the baseline score as a covariate and with the raw score as the dependent variable. Since the two techniques yielded similar trends, the analysis of change scores is presented here. The one way ANOVA was combined with the *a posteriori* Tukey test in order to isolate significant differences (Scheffe, 1959).

In addition to the overall analyses among drug groups, patients were stratified into a cognitively "non-impaired" group - those with scores lying within the normal range on TMB, and an "impaired" group - those with scores lying below the normal range. The cutoff score employed to divide the two groups was 92 seconds, based on norms provided by Golden et al (1980). The Kruskal-Wallis K sample test was used to test for differences in TMB scores among TMB-impaired patients in the three treatment groups over the entire 24 week study period. Differences from baseline to Week 24 and from Week 8 to Week 24 within each treatment group were assessed using the Wilcoxon Matched-Pairs Ranked-Signs test.

Results

Of the 761 patients enrolled, 620 participated in baseline evaluations and were entered into one of the three treatment groups. Demographic and clinical characteristics of the patients in the three groups at baseline were comparable (Croog et al, 1986). The most common reason for patients not entering active therapy was failure to meet the required blood pressure cri-

Changes in Mean Scores on the TMB from Baseline to Week 24 by Treatment Group*

	N	Baseline	Week 24	% Change	Compariso.. Between Groups
Captopril (A)	104	97.9 ± 4.4	76.9 ± 2.7	-14.2 ± 2.9	
Methyldopa (B)	85	95.6 ± 5.3	84.8 ± 4.4	- 4.9 ± 3.3	A–B**
Propranolol (C)	110	91.3 ± 3.9	77.0 ± 3.0	-10.2 ± 2.9	

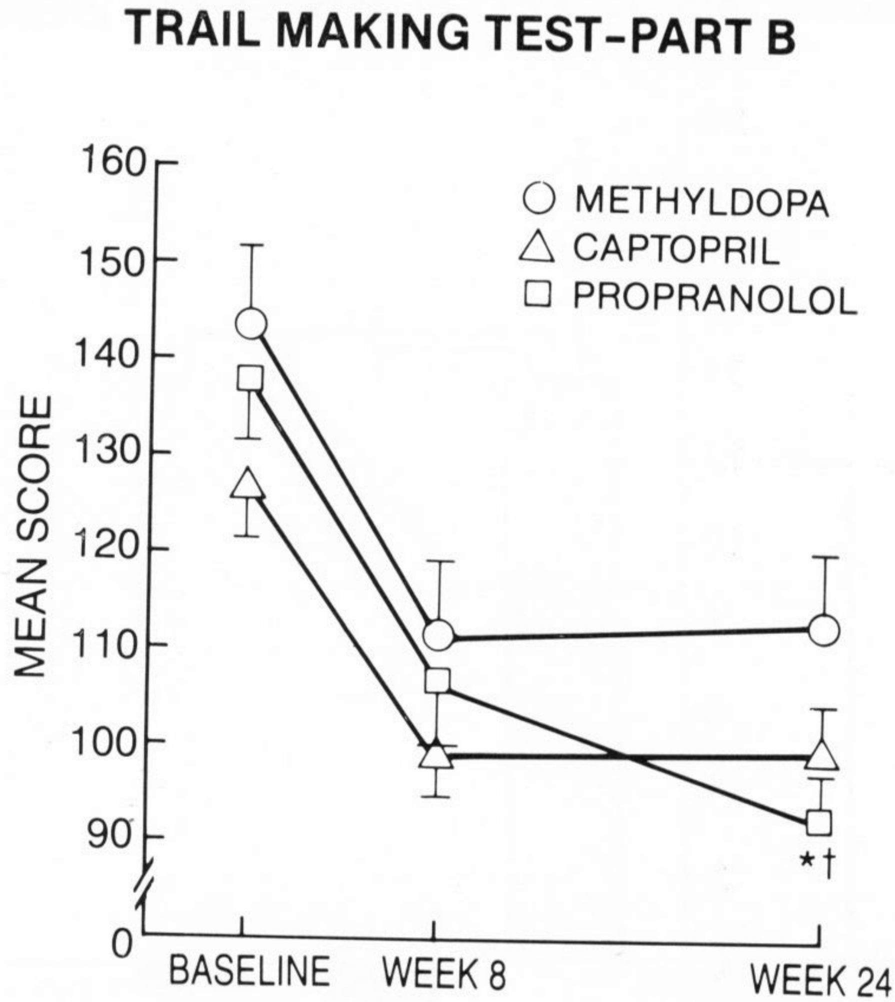
*A decreasing score denotes improvement.

**$p < 0.05$

Demographic and Clinical Variables for TMB Impaired and Non-Impaired Patients in the Three Groups at Baseline*

	Non-Impaired			Impaired		
Variable	Captopril (N = 59)	Methyldopa (N = 53)	Propranolol (N = 63)	Captopril (N = 45)	Methyldopa (N = 32)	Propranolol (N = 47)
Demographic						
Age at entry (yr)	43.4	47.4	44.6	51.4	51.8	48.0
Education (median school yr completed)	15	16	15	14	12	14
Occupation (%)						
Executive/professional	44.1	36.5	36.5	23.2	15.6	14.9
Small business/administrative	20.3	30.8	31.8	30.2	31.2	23.4
White collar/clerical	13.6	9.6	12.7	20.9	18.8	25.5
Blue collar	22.0	23.1	18.8	25.7	34.4	36.2
Median income ($)	33,900	35,600	34,200	33,300	35,000	32,750
Percent married	79.7	77.4	82.5	77.8	90.6	89.4
Clinical						
Duration of essential hypertension (yr)	7.0	5.9	6.6	5.5	8.1	7.1
Weight (Kg)	90.5	89.5	93.7	91.6	88.8	91.6
Systolic blood pressure (mm Hg)	140.0	142.9	142.9	145.3	141.3	140.1
Diastolic blood pressure (mm Hg)	96.1	96.3	96.6	97.0	96.9	96.2
Trail Making B score	67.8	66.8	64.8	137.4	143.1	126.9

*Data reported are means unless otherwise specified.



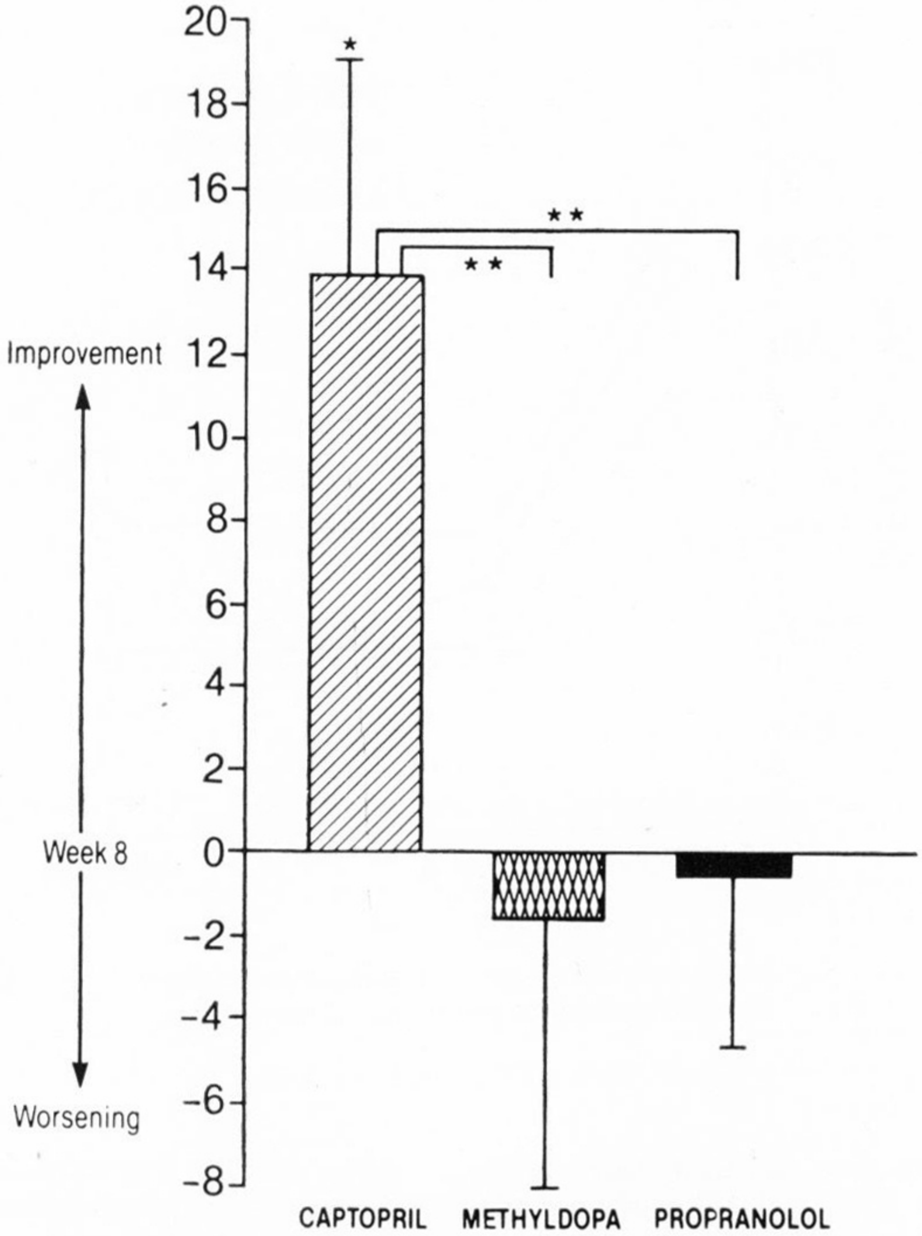
* percentage change from baseline different from that for methyldopa or propranolol, $p < 0.05$

† different from methyldopa, $p < 0.02$

teria. One hundred and thirty-four (22%) of the above 620 patients discontinued participation at sometime prior to completion of the study. The main reason for discontinuation from active therapy were adverse reactions (N = 83) and loss to follow-up (N = 27). Other reasons were unacceptably high diastolic blood pressure, concomitant illness, non-adherence to protocol requirements, and poor compliance. The distribution by treatment group of patients who discontinued was 31 (15 %) for captopril, 54 (27 %) for methyldopa, and 49 (23 %) for propranolol. The rate of withdrawal with captopril was significantly lower ($p < 0.05$) than with either methyldopa or propranolol.

After visit B8, 178 of the remaining 486 patients required the addition of diuretic medication for blood pressure control and, as mentioned earlier, were excluded from analyses. Thus, a total of 308 patients completed the 24-week active treatment period on monotherapy and, with the exception of nine patients from whom data on cognitive tests was incomplete, comprise the data base for this report. No significant differences were found among treatment groups with regard to baseline characteristics and cognitive scores. Comparison of cognitive scores at baseline, week 8, and week 24 demonstrated a general improvement over time in all groups. Such improvement may be expected, as reported by

CHANGE IN TMB SCORES FROM WEEK 8 TO WEEK 24

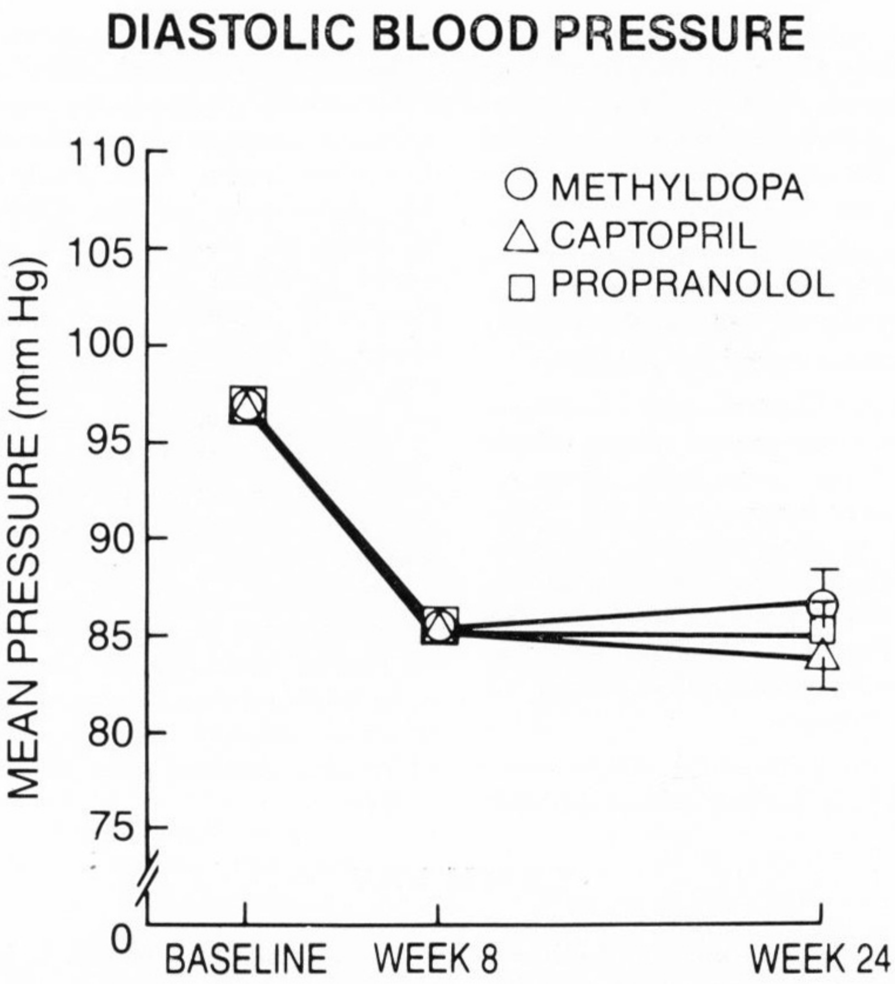


* $p<0.01$. statistical contrast of 8-24 week change compared to 0

* * $p<0.05$. statistical test for pairwise comparison

Craddick and Stern (1963), as a result of increased familiarity with the test on repeated testing (i.e., practice effect).

Comparison of Change Scores Between Drug Groups: Of primary interest, of course, is the question of possible differences in degree of improvement in cognitive test scores among drug groups. Comparisons among the three treatment groups baseline to visits B8 and B24 suggested no significant difference in overall mean change on the VRT or the TMA. However, as shown in Table 1, the magnitude of improvement in mean TMB scores after 24 weeks of treatment was greater for captopril than for propranolol or methyldopa. The difference was statistically significant against methyldopa ($p<0.05$).



Changes in Scores in Patients with Differing Levels of Cognitive

Performance: As noted previously, the patient population was stratified according to scores on the TMB at baseline, with those scoring 92 seconds or higher identified as "impaired" and those scoring less than 92 seconds as "non-impaired". This stratification was undertaken to determine the effect of treatment acording to the level of cognitive functioning at baseline.

Characteristics of the impaired and non-impaired sub-populations at baseline are shown in Table 2. As can be seen across treatment groups, impaired patients were older, had less formal education, and were more likely to be white and blue collar workers than were non-impaired patients. As shown in the table, patients were quite similar across treatments within both the impaired and non-impaired groups. No significant differences across treatment groups existed on baseline TMB scores among either impaired or non-impaired patients.

While no significant differences occurred over the entire 24-week study period either within or between drug groups among TMB-non-impaired patients, changes in TMB scores among TMB-impaired patients in the three treatment groups did occur and are shown in Figure 7. By the end of the study, those patients that received captopril had improved significantly more ($p < 0.05$) in performance on the TMB than those receiving either methyldopa or propranolol. The difference in mean scores was significant ($p < 0.02$) between captopril and methyldopa at week 24.

No significant differences across the groups were found during the first eight weeks of active therapy. Hence, the differences for the 24-week period reflect mainly changes during the period following the first eight weeks. Thus, Figure 8 illustrates the changes in TMB scores for the three drugs during the final 16 weeks of treatment using scores at week 8 compared with scores at week 24. TMB-impaired patients on captopril improved significantly ($p < 0.01$) while those on propranolol or methyldopa showed slight declines in performance. The differences between captopril and either of the comparison drugs were also significant ($p < 0.05$).

These differences were sustained when scores on the TMA were subtracted from scores on the TMB to reduce the component of psychomotor speed in the latter.

Blood Pressure Control and Cognitive Function Scores: Comparisons of the effects of the effects of the three study drugs on blood pressure were made within the TMB-impaired group to determine the differential cognitive effects found were secondary to differential blood pressure response. As can be seen in Figure 9, blood pressure was found to be similarly controlled within all three treatment groups.

In summary, no differential effects occurred on the VRT, a finding consistent with results of earlier studies (Franceschi et al, 1982; Solomon et al, 1983), or on the TMA, which is generally regarded as providing a measure of psychomotor speed rather than higher level cognitive processes. The differences on the TMB favored the inhibitor of ACE captopril and were restricted primarily to a sub-group of patients those with relatively poor cognitive performance at baseline. The differential cognitive effects among impaired patients do not appear to have resulted from variation in antihypertensive effects of the three drugs. Indeed, blood pressure was similarly controlled among impaired patients in each treatment group. On the other hand, while a "regression toward the mean" effect or the occurrence of practice effects on repeated testing could explain the overall tendency to improvement during the first 8 weeks of active therapy, neither can explain the differences obtained after the subsequent 16 weeks.

SUMMARY

Results from both preclinical and clinical studies described here suggest that ACE may have a role in the modulation of cognitive memory processes in the rat and in humans. The finding of improved cognitive performance among patients treated with captopril relative to those treated with propranolol or methyldopa is consistent with other clinical and prec-clinical data. Clinical data derive primarily from quality of life measures based on interviews with patients in the same clinical trial from which our other cognitive data are drawn. For example, mental acuity in the workplace was reported to have improved significantly from baseline to week 24 in patients on captopril ($p<0.05$), although it did not change in patients treated with propranolol and worsened in those receiving methyldopa (Croog et al, 1987). The difference between captopril and methyldopa was significant ($p<0.01$). Pre-clinical data come primarily from studies demonstrating that inhibitors of ACE delay CAE in rats when compared not only with methyldopa, but also with saline (Sudilovsky et al, 1984, 1986).

A fundamental question is how could inhibition of ACE improve cognitive functioning independent of blood pressure control. It is known that captopril exerts its antihypertensive effects primarily through inhibition of the ACE and that this is present in the brain as well as in non-neuronal tissues elsewhere (Ganten et al, 1982; Strittmatter et al, 1983, 1984). The activity of the enzyme has been found to be significantly increased in the caudate nucleus, the frontal cortex, parahyppocampal gyrus, and medial hippocampus of patients dying with Alzheimer's disease when compared to age-matched controls (Arregui et al, 1982). In addition, AII has been shown to impair performance on various learning and memory paradigms in animals (Melo and Graeff, 1975; Morgan and Routtenberg, 1977). Raising the level of endogenous AII by intravenous administration of its precursor renin has similar effects, and these are prevented if captopril is administered previously (Koller et al, 1979). Also, chronic oral treatment with captopril produces changes of brain renin angiotensin system parameters which suggest inhibition of AII biosyntheses in the brain (Scholkens et al, 1983). It is conceivable therefore, that our findings with prolonged administration of captopril are exerted through reduced formation of AII.

Other possible mechanisms through which captopril may affect cognition include peptide-protein systems (Ganten et al, 1982, Sudilovsky et al, 1988) or the modulation of cerebral blood flow autoregulation mechanisms. In hypertensive rats it has been shown that captopril protects against the effects of cerebral ischemia (Capdeville et al, 1986). Following its intravenous administration the lower and upper limits of cerebral blood flow autoregulation are reset to lower mean arterial pressure than in controls (Barry et al, 1984). In man, captopril has a similar effect on cerebral blood flow autoregulation (Paulson et al, 1985). This may explain the observed absence of signs of neurological impairment with very low blood pressure as well as the restoration of cerebral blood flow after captopril in patients with congestive heart failure (Barry et al, 1984; Rajgopalan et al, 1984).

Whatever the mechanism of action might be, the empirical data reported here suggest that inhibition of the ACE may offer a new avenue for the treatment of cognitive and memory deficits which merits further exploration with other animal models and in controlled studies involving appropriate patient populations.

RESUMEN

Los Resultados de los estudios preclínicos y clínicos presentados sugieren que la ACE (Angiotensine Conversion Enzyme) puede tener implicancia en la modulación de los procesos de memoria cognitiva en la rata y en el hombre. La constatación del mejoramiento de las realizaciones en los pacientes tratados por el captopril (inhibidos de la ACE) en comparación con aquellos tratados por el propranolol o la metildopa, se aprecia que está de acuerdo con otros resultados preclínicos (retardo de CAE por los inhibidores de la ACE) y clínicos (medidas de la calidad de la vida y de la eficacia profesional).

El problema es de saber como la inhibición de la ACE puede mejorar las funciones cognitivas independientemente del control de la presión arterial. Se sabe que el captopril debe esencialmente su acción antihipertensiva a la inhibición de la ACE y que ésta se ejerce tan bien al nivel del encéfalo como de los tejidos extraños al sistema nervioso. La actividad de la enzima ha sido hallada significativamente aumentada en el núcleo caudado, la corteza frontal, el giro parahipocámpico y el hipocampo de pacientes muertos de enfermedad de Alzheimer, en comparación con testigos de la misma edad. Otros posibles mecanismos de la acción del captopril sobre las funciones cognitivas comprenden sistemas péptidos-proteínicos o la modulación de los mecanismos de auto-regulación de la circulación cerebral.

Cualquiera que sea el mecanismo, los resultados empíricos que se han mencionado, sugieren que el inhibidor de la enzima de conversión pueda abrir una nueva vía a los tratamientos de los déficits cognitivos y nmésicos. Merecen nuevas investigaciones con otros modelos de animales y estudios controlados que engloban adecuadas poblaciones de enfermos.

RÉSUMÉ

Les résultats des études précliniques et cliniques présentées suggérent que l'ACE (Angiotensine Conversion Enzyme) peut avoir un rôle dans la modulation des processus de mémoire cognitive chez le rat et chez l'homme. La constatation d'améliorations des performances chez les patients traités par le captopril (inhibiteur de l'ACE) comparativement à ceux traités par le propranolol ou la méthyldopa s'accordent avec d'autres résultats précliniques (retard de CAE par les inhibiteurs de l'ACE) et

cliniques (mesures de la qualité de la vie et de l'efficacité professionnelle).

La question est de savoir comment l'inhibition de l'ACE peut améliorer les fonctions cognitives indépendemment du contrôle de la pression artérielle. On sait que le captopril doit essentiellement son action antihypertensive à l'inhibition de l'ACE et que celle-ci s'exerce aussi bien au niveau de l'encéphale que des tissus étrangers au système nerveux. L'activité de l'enzyme a été trouvée significativement augmentée dans le noyau caudé, le cortex frontal, le gyrus parahippocampique et l'hippocampe de patients morts de maladie d'Alzheimer, en comparaison avec des témoins de même âge. D'autres mécanismes possibles de l'action du captopril sur les fonctions cognitives comprennent des systèmes peptido-protéiniques ou la modulation des mécanismes d'auto-régulation de la circulation cérébrale.

Quelque soit le mécanisme, les résultats empiriques ici rapportés suggérent que l'inhibiteur de l'enzyme de conversion peut ouvrir une voie neuvelle au traitements des déficits cognitifs et mnésiques. Ils méritent de neuvelles investigations avec d'autres modèles animaux et des études contrôlées englobant des populations adéquates de malades.

ZUSAMMENFASSUNG

Die Ergebnisse der praeklinischen und klinischen Studies lagen nahe, dass das ACE (Angiotensine Conversion Enzyme) auf die Modulation des kognitiven Gedaechnisses Einfluss haben kann, bei der Ratte und dem Menschen.

Die Feststellung der Besserungen der Realisationen bei Menschen, die mit Captopril (einem ACE-Hemmer) behandelt wurden, verglichen mit jenen, die mit Propanool oder methyldopa behandelt wurden stimmen ueberein mit anderen praeklinischen (Verzoegerung desCAE durch die ACE-Hemmer) und klinischen Resultaten (Abschaetzung der Lebensqualitaet und der beruflichen Effizienz).

Esbandelt sich darum, zu wissen wie die ACE-Hemmung die Kognitiven Funktionen bessern kann unabhaengig von der Kontrolle des Blutdrucks. Man weiss, dass Captoprilseine antihypertensive Wirkung wesentlich der ACE-Hemmung verdankt, und dass dies sowohl von Gehrim als auch den Geweben ausserhalb des Nervensystems ausgeuebt wird.

Die Wirkung des Enzyms ist wesentlich vermehrt in Nucleus cadatus, in Gyrus parahippocampi und im Hippocampus von Patienten, die an Alzheimerscher Krankbeit gestorben sind, gegenueber anderen Kranken gleichen Alters Andere moegliche Wirkungsmechanismen des Captoprils auf die kognitiven Funktionen sind Systeme vonPeptiden-Proteinen oder dieModulation der Autoregulationsmechanismen des Gehornblutkreislaufes.

Welches auch immer der Mechanismus sei, die erwaehnten empirischen Resulate legen nahe, dass der Hemmer des konversios-enzyms neue Wege oeffnen kann zu den Behandlungsmoeglichkeiten des kognitiven und mnesischen Defizits, Sie Berechtigen zu neuen Untersuchungen mit anderen Tiermodellen und Untersuchungen mit geeignenten Populationen.

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LONG-TERM POTENTIATION AND MEMORY

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INTRODUCTION

In 1973, two reports appeared in the *Journal of Physiology* that described a Long-Term Potentiation (LTP) of synaptic transmission at a hippocampal monosynaptic junction in the mammalian CNS (Bliss and Gardner-Medwin, 1973; Bliss and Lomo, 1973). These papers were of considerable interest because they marked the first demonstration of an alteration in synaptic efficacy in the mammalian brain which possessed a lengthy time-course. The changes (on the order of a 50-250% increase in the amplitude of the evoked population response) lasted for at least 10 hours in the acute anesthetized preparation and up to sixteen weeks in the chronic unanesthetized preparation following a series of tetanic stimuli delivered to the afferent fibers. These results attracted great interest because of the possibility that the phenomenon might underlie some aspects of memory storage.

Long-term potentiation is defined as a stable, relatively long lasting increase in the magnitude of a post-synaptic response to a constant afferent volley following brief tetanic stimulation of the same afferents (Fig. 1). LTP can also be observed to occur in conjunction with the learning of a behavioral task — in the absence of tetanic stimulation. Initially observed in the hippocampus, LTP has now been documented in a variety of brain structures and in a variety of species (for a review, sea Teyler & DiScenna, 1987). This paper reviews the phenomenology of LTP and its hypothesized mechanism of action.

The Distribution of LTP in the Brain

LTP was initially observed in the intrinsic hippocampal synapses (areas CA1, CA3 and dentate) and more recently has been documented in extrinsic hippocampal synapses, other limbic synapses, medial geniculate nucleus, pyriform cortex, primary visual and somatosensory cotex and cerebellar cortex and deep nuclei (see review by Teyler & DiScenna, 1987).

The ontogenetic development of LTP has been examined in the rodent hippocampus, and neocortex where it is found to develop between postnatal days 7 and 10 for area CA1 (Harris & Teyler, 1984) and visual cortex (Perkins & Teyler, 1987) and somewhat later (7-28 days) in the dentate gyrus (Wilson, 1984). The magnitude of LTP peaks at about day 15, falling subsequently to adult levels. LTP thus appears to be a widespread phenomenon, however, it is not yet established that these diverse examples of enhanced synaptic activity all represent the same phenomenon. In many

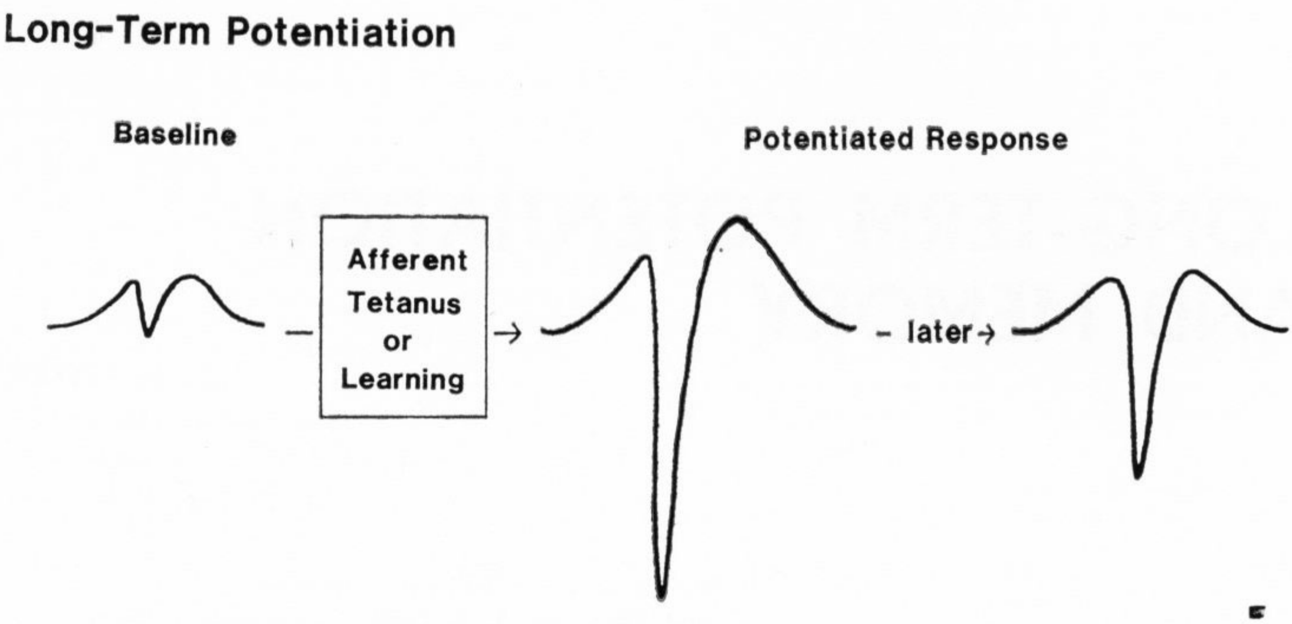

Fig. 1. — *Long-Term Potentiation (LTP) is an enduring change in synaptic efficacy seen at synaptic junctions of the mammalian CNS. Shown here as an increase in the size of a hippocampal field potential following either electrical tetanization of afferents or a behavioral learning experience, LTP gradually decays back to baseline.*

experiments the parameters required to initiate LTP (as well as the phenomenon of LTP itself) are different than the hippocampus. The question of whether all of these forms of plasticity represent a unitary phenomenon or whether different phenomena are grouped under the heading of LTP will ultimately be answered by determining the mechanism underlying LTP.

Features of LTP

The experimental induction of LTP begins with the delivery of tetanic stimulatin to afferent fibers. LTP can be induced by a wide range of tetanus frequencies and patterns. Tetanus frequencies supporting LTP range from 0.2 Hz to 400 Hz a total of 100-200 pulses delivered. Following termination of the tetanus it is possible to record a substantial post-tetanic potentiation (PTP) from the tetanized synapses (Bliss & Lomo, 1973; Harris & Teyler, 1984) prior to the development of LTP.

The duration of LTP is considerable by neurophysiological standards, but insufficient when considered in behavioral terms. The original descriptions of LTP in intact rabbits indicated a duration of about three days following a single tetanus (Bliss & Gardner-Medwin, 1973). However, they also showed that the duratin could be extended considerably by repeating the tetanic stimulation. Comparable studies on rodents have demonstrated decay rates ranging from three days to 13 days (Barnes, 1979; Racine et al, 1983).

LTP displays input specificity (also termed homosynaptic potentiation) such that only those inputs that are simultaneously active (or nearly so) can be potentiated (Andersen et al, 1977; Lynch et al, 1977). LTP thus appears to be a local process and not a generalized increase in neuronal excitablility. Intracellular studies of LTP do not reveal constent changes in membrane potential or input resistance (Andersen et al, 1980; Barrionuevo & Brown; 1983). Input specificity suggests that some of the mechanisms subserving LTP of the population EPSP should be located near the activated synaptic membrane.

A stimulus intensity threshold exists for the production of LTP at the perforant path-dentate granule cell synapses. This LTP threshold is at or near the stimulus intensity required to elicit a population spike (McNaughton et al, 1978). The population spike, per se, does not appear to be necessary for LTPP, however, rather, it appears that a minimum number of afferent fibers (or synaptic contacts) must be coactive in order for LTP to occur. This probably reflects a requirement for a given level of depolarization in the postsynaptic cells.

Related to the coactivation phenomenon is the associative LTP paradigm. In associative LTP the concurrent activation of independent weak and strong synaptic inputs produce LTP of the weak input, where tetanization of the weak input above would not result in LTP. (Levy & Steward, 1979; Robinson & Racine, 1982; Barrionuevo & Brown, 1983.

Interest in associative LTP derives primarily from the potential for such a phenomenon to facilitate associations between coative elements on a target neuron. D. O. Hebb (1949) proposed that correlated afferent activity and postsynaptic activation leads to an alteration in synaptic efficacy. Since LTP can be induced in the absence of action potentials (McNaughton et al, 1978) in the postsynaptic cell, it is necessary to modify the "Hebb Synapse" to a system which requires coactivity among converging presynaptic elements onto a common postsynaptic membrane sufficient to achieve a supra-LTP threshold.

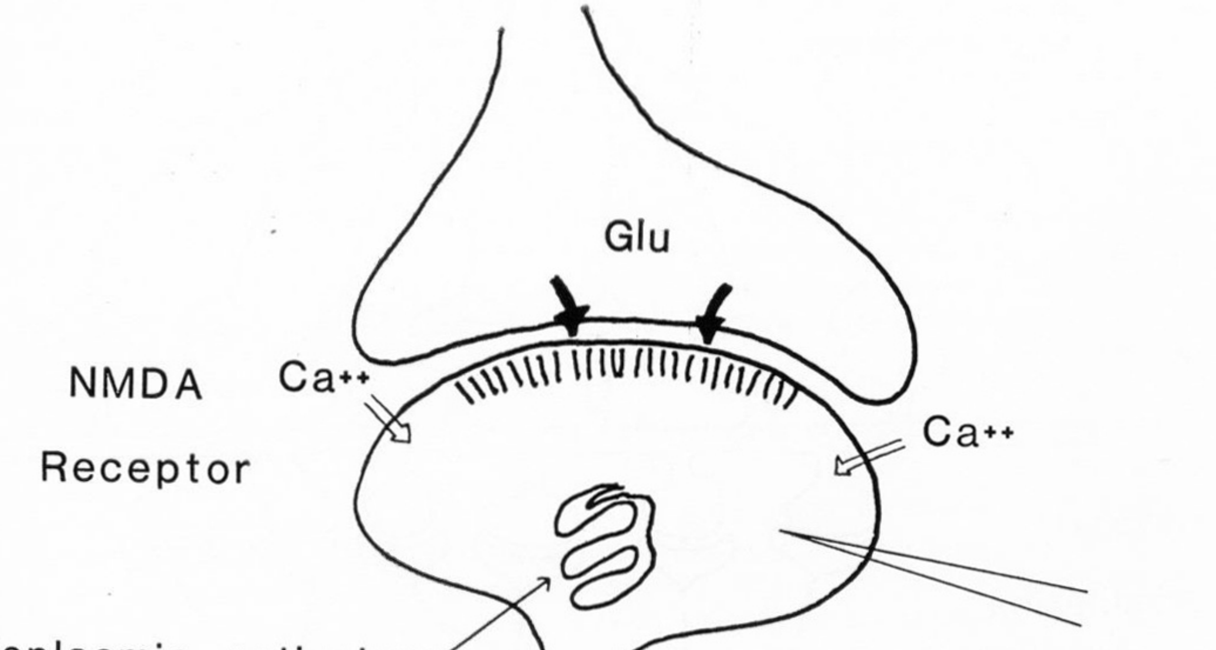

Fig. 2. — *The NMDA receptor (a subtype of glutamate receptor) has been implicated in the cellular mechanism of LTP. Normally blocked by physiological concentrations of Mg+2 (and thus not involved in normal synaptic transmission) the NMDA receptor is activated by depolarization (as induced by tetenic presynaptic stimulation) resulting in the opening of Ca+B channels. The increased ca+B conductance is believed to be responsible for the triggering of the enduring aspects of LTP, perhaps acting through Ca+B sequestering organelles such as he endoplasmic reticulum. LTP can be induced without tetanic presynaptic stimulation by concurrent postsynaptic depolarization and synaptic activation or glutamate application.*

Neuronal Mechanisms Underlying LTP

Synaptic transmission is required for LTP induction. This has been demonstrated with the use of neurotransmitter antagonist and manipulation of Ca+2 concentration. Given the central role of calcium in synaptic processes, several laboratories have studied the role of calcium-calmodulin systems in LTP. Dunwiddie et al. (1978) and Wigström et al (1979) showed that LTP was blocked under low calcium conditiones, indicating that calcium plays a critical role in LTP production. The experimental paradigm involves delivering the tetanic stimulus in a low calcium environment (all experiments done *in vitro*) and testing for LTP later in a normal calcium environment. These observations were extended by the experiments of Baimbridge and Miller (1981), who demonstrated that the uptake and retention of labelled calcium is increased after LTP. Kuhnt et al (1985) demonstrated a large increase in dendritic calcium deposits with the induction of LTP.

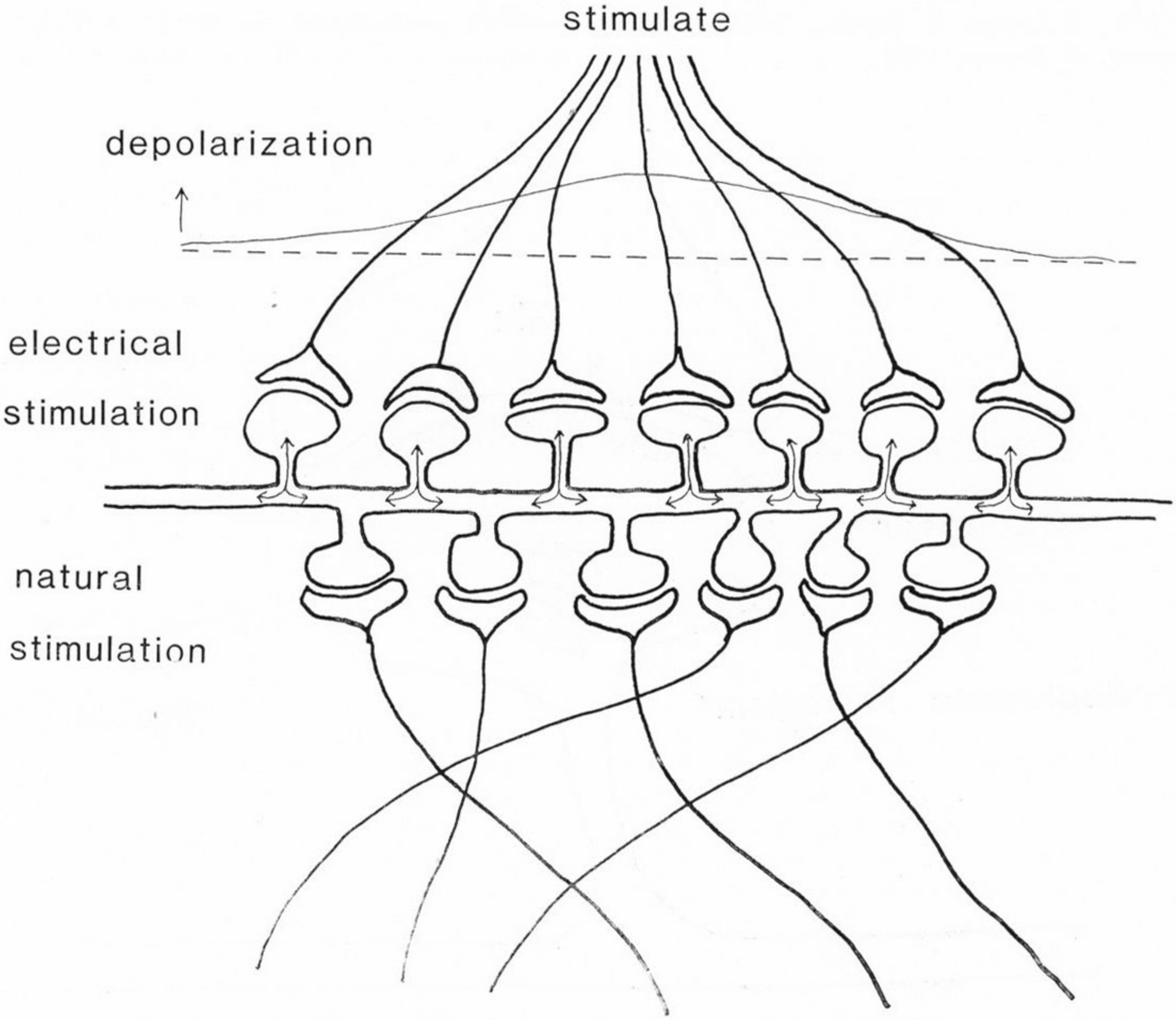

Fig. 3. — *Electrical stimulation of afferents (top) is thought to synchronously activate spines over a limited extent of the dendritic tree - an event rarely encountered in nature. The depolarizing currents so produced summate between adjacent spines resulting in a segment of dendrite sufficiently depolarized to override the voltage-dependent, Mg+2–mediated, blockade of Ca+2 conductance. In nature (bottom) it is hypothesized that co-active synapses, arising from diverse sources, also display the capacity to activate the NMDA channel. This latter property is the basis of the associational capabilities of LTP - the ability to interrelate elements that are co-active and nearly coterminus.*

Since glutamate is believed to be the neurotransmitter at the principal trisynaptic junctions of the hippocampus (White et al, 1977), Dunwiddie et al (1978) tetanized perforant path fibers while bathing the dentate gyrus slice in APB (a glutamate antagonist). No LTP was later observed after washout of the APB, suggesting that glutamate neurotransmission is necessary for LTP. Three subvarieties of the glutamate receptor are known to exist: Kainate, Quisqualate and N-methyl-D-aspartate (NMDA). Recently, there has been considerable interest in the activity of the NMDA receptor (Fig. 2; Collingridge & Bliss, 1987). A specific NMDA antagonist, D-2-amino-5-phosphonovalerate (APV) does not affect normal transmission (which appears to be gated through kainate/quisqualate receptors), but it does block the experimental induction of LTP (CA1, slice preparation Collingridge et al, 1983; Wigström & Gustafsson, 1984 Harris et al, 1984), as well as behavioral LTP (Morris et al, 1986).

During normal activity, MNDA receptors appear to be blocked by a voltage dependent, Mg+2– mediated blockade (Nowak et al, 1984). The depolarization produced by the tetanic stimulus alleviates the MG+2 block and opens the MNDA receptor channels. The NMDA channel has a high conductance to Ca+2, resulting in a build-up of intracellular Ca+2 concentrations (Fig. 3). This Ca+2 can then activate a) Ca+2– dependent K+ or C1– conductances (MacDermott & Dale, 1987), b) Calpain, a Ca+2– dependent protease which unmasks/activates a quiescent population of glutamate receptors (Lynch & Baudry, 1984), c) Ca+2– activated second messenger systems (Collingridge & Bliss, 1987), and/or d) mechanisms utimately responsible for synaptogenesis (Chang & Greenough, 1984).

Lynch et al (1983) showed that intracellular injection of the calcium chelator, EGTA into area CA1 pyramidal cells (hippoocampal slice) blocks the appearance of LTP. The most startling result is that of Turner et al (1982) who demonstrated that merely incubating a hippocampal slice for five to ten minutes in medium containing elevated calcium levels (4 mM) is sufficient to produce LTP that is identical in many ways to that produced by afferent activation. Considering the ubiquitous role of calcium in neuronal regulation it is not surprising that manipulations of calcium levels would influence LTP, but these results suggest that there is an additional role of calcium in LTP beyond that of normal synaptic transmission.

Behavioral LTP

Although LTP has been known for over a decade, research into its behavioral correlates is recent. Much of the interest in LTP relates it to the possibility that LTP may be involved in memory storage. To this end it is essential that it be convincingly demonstrated that LTP is produced in association with behavioral learning in intact animals and is necessary and sufficient for the behavioral learning. Among the first studies of the behavioral correlates of LTP was that of Barnes (1979) who studied the behavioral and electrophysiological properties of young and old rats. She demostrated a positive relationship between the longevity of electrically induced LTP and the level of retention on a behavioral task.

In an attempt to determine if LTP occurs during the normal activity of an animal, as opposed to a laboratory learning task, dentate gyrus synapses were studied in chrinically implanted rats who were placed in a novel, complex environment (Sharp et al, 1985). No tetanic stimuli were delivered, only a single daily probe stimulus. Across several days in the novel environment an LTP-like change was observed, which gradually subsided, only to be reinstated upon transfer to a second, different complex environment. An important controlused in this study was to place animals in restricted environments for 1 month so that any prior behavioral LTP might decay to baseline before the experimental session. Greenough and colleagues (Black & Greenough, 1986) have demonstrated that exposure to an enriched environment, even briefly, results in morphological changes at the synaptic level, perhaps suggests a role for experience-dependent synaptogenesis in neuronal plasticity.

In a more direct test of the behavioral correlates of LTP, Thompson and colleagues chronically implanted rabbits with electrodes to monitor dentate gyrus synapses. The rabbits were trained on a classical conditioned eyeblink (nictiating membrane) response (Thompson et al, 1983). Since no tetanic stimuli were presented, any change in dentate synaptic efficacy must be elicited by naturally occurring patterns of afferent activity. The results indicated that dentate synaptic efficacy increased approximately in parallel with the acquisition of the behavioral response (Weisz et al, 1984). Similar results showing that behavioral training results in LTP like changes at monosynaptic junctions in hippocampus have been reported by Laroche an Bloch (1982) in a tone-footshock conditioning paradigm and by Ruthrich et al (1982) in a brightness discrimination task.

Disterhoft et al (1986) obtained evidence that beravioral eyelid conditioing of rabbits subsequently results in a Ca+2-mediated reduction in K+ current of hippocampal CA1 cells studied *in vitro*. The demostration that neuronal changes induced *in vivo* can be preserved and studied *in vitro* is important and may permit an extension of the advantages inherent in the brain slice preparation to beravioral studies.

These results, taken together, provide some evidence that behavior proceses give rise to LTP-like phenomenon which are similar in many respects to LTP induced by electrical stimulation.

LTP Effects on Learning

If LTP has a relationship to the neuronal mechanisms underlying behavioral learning, then the previous establisment or blockade of LTP in circuits responsible for behavioral responding should alter the behavior in some way. Clearly one could hypothesize that the induction of LTP would either facilitate or disrupt subsequent behavioral responding depending upon the effect of the induced LTP on the signal processing characteristics of the potentiated circuit. Berger (1984) investigated this question in the rabbit nictitating membrane conditioning paradigm. Electrical stimulation was used to produce LTP in the dentate gyrus of untrained rabbits. Animals were then trained on the eyelid response. Animals receiving LTP treatments prior to training subsequently showed accelerated behavioral learning in this paradigm. Morris et al (1986) demonstrated that the NMDA antagonist AP5 blocked the behavioral learning of a spatial task, further implicating the NMDA receptor in neuronal plasticity. However, many more experiments are needed in the area of behavioral LTP in order to be confident of the nature of the relationship between the beravioral learning and LTP.

Conclusions

The existence of a neural phenomenon that persists for considerable lengths of time and is seen to occur both as a result of electrical stimulation of afferents and in conjunction with behavioral learning has led many to consider the hypothesis that LTP underlies long-term information storage in the mammalian brain (Goddard, 1980 Eccles, 1983; Lynch & Baudry, 1984; Teyler & DiScenna, 1984; 1987). A sufficiently robust test of this hypothesis will require a better understanding of the neuronal mechanisms underlying LTP, particularly the role of Ca+2 in the maintenance of LTP, and a clearer appreciation of the links between LTP and behavioral learning.

Acknowledgements

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SUMMARY

Long-term potentiation is an enduring alteration in monosynaptic eficacy seen in a variety of synaptic junctions in the mammalian central nervous system. First discovered in 1973, LTP has been extensively studied both in terms of its underlying mechanism of action and more recently in terms of its behavioral significance. LTP is known to be input specific and to obey a modified "Hebbian" rule. Investigations into the cellular mechanisms underlying LTP have show that LTP is correlated with increased glutamate - release, increased glutamate receptor binding, alterations in the morphology of synaptic contacts, and involvement with the NMDA receptor. Behavioral studies show that LTP can be elicited as a result of behavioral learning experiences and that the prior induction of LTP by electrical stimulation can either facilitate or impare subsequent behavioral learning, depending upon the task conditions. A hypothesis will be examined that long - term potentiation in hippocampus anad neocortex may subserve information storage in the central nervous system.

RESUMEN

La potencialización para largo plazo (LTP) es un cambio facilitado y prolongado de la eficacia monosináptica que puede ser observada a nivel de distintas uniones monosinápticas del sistema nervioso del mamífero. Descubierto en 1973 LTP fue muy estudiado tanto en lo relativo a la razón de su mecanismo de acción y mas recientemente, en lo que corresponde a su significación comportamental. LTP es conocido por responder a todo estímulo sináptico específico y obedece a la ley de Hebb modificada. Investigaciones relativas a la comprensión de los mecanismos celulares basales en la LTP evidenciaron que esta LTP está en relación con el aumento de liberación de glutamato y la ocupación de los asientos receptores de glutamato, así como de los cambios morfológicos de las uniones sinápticas y también en relación con el receptor de NMBA. Estudios comportamentales demuestran que la LTP puede ser promovida a continuación de experiencia de aprendizaje de conducta y que toda LTP inducida previamente por estimulación eléctrica, puede o bien facilitar o disociar un susecuente aprendizaje del comportamiento y esta en condiciones de trabajo muy particulares.

RÉSUMÉ

La potentiation à long terme ("LTP") est un changement facilité et prolongé de l'efficacité monosynaptique qui peut être observée au niveau de diverses jonctions monosynaptiques du système nerveux mammalien. D'abord découverte en 1973, LTP fut longuement étudiée à la foies en ce qui regarde la raison de son mécanisme d'action et, plus récemment, en terme de sa signification comportementale. LTP est connue pour repondre à tout spécifique stimulus et obéit à la loi de Hebb modifiée. Des investigations relatives à la compréhension des mécanismes cellulaires à la base de la LTP ont montré que cette LTP est en relation avec l'augmentation de libération du glutamate et l'occupation des sites récepteurs à glutamate, ainsi que des altérations morphologiques des jonctions synaptiques et aussi en relation avec le récepteur à MNDA. Des études comportementales démontrent que la LTP peut être élicitée à la suite d'expérience d'apprentissage de comportement et que toute LTP, induite au préalable par stimulation électrique, peut ou bien faciliter ou dissocier subséquent apprentissage de comportement et ce, dans des conditions de travail bien particulières.

ZUSAMMENFASSUNG

Die Langzeitpotenziation (LTP) ist eine bahnende und langandauernde Aenderung der monosynaptischen Wirksamkeit, die man bei den verschiedenen monosynaptischen Kontakten im Nervensystem der Saeugetiere beobachten kann. Die 1973

entdeckte LTP wurde bezueglich des Wesens sihres Mechannismus und neuerdings auch bezueglich ihrer Bedeutung fuer das Verhalten intensiv untersucht. LTP ist bekannt als Reaktion auf jjedden spezifischen Reiz und folgt dem modifizierten Hebbschen Gesetz. Untersuchungen fuer das Versstaendnis der grundlegenden baszelylulaeren Mechanismen der LTP bewiesen, dass diese LTP in Verbindung mit der Produktionsverstaerkung von Glutaman und der Besetzung von Glutamant Rezeptoren steht, sowie auch mit den morphologischen Veraenderungen der synaptischen Beruehrungsstellen und uch in Verbindung stehen mit dem NMBA - Rezeptor. Verhaltungsstudien bezeugen, dass LTP hervorgerufen werden kann durch die Erfahrung des Erlernens des Verhaltensweisen und dess jejde LTP, die vorther durch elektrische Reizung hervorgerufen ist, eine darauffolgende Erlernung der Verhaltungsweise bahnen oder diessoziieren kann, und unterganz besonderen Arbeitsbedingungen steht.

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