ly immunosuppressed patients. Nevertheless, the results of our experiments, which strengthen the evidence of autoimmune factors in diabetes, indicate that recurrent disease could be a potential problem, especially if optimal histocompatibility, improved drugs, or other protocols in the future allow avoidance of rejection with no or minimal immunosuppression. A precedent for such a problem is provided by the results of renal transplantation in victims of another, probably autoimmune, disease, glomerulonephritis. Recurrence of the original disease occurred in normal kidneys taken from identical twins in 11 of 17 instances (*13*).

The finding that neonatal BB rats inoculated with WF bone marrow cells failed to develop the expected incidence of diabetes in later life suggests an entirely new approach to therapy or prophylaxis of diabetes. The mechanism by which allogeneic bone marrow protected these rats from diabetes is not obvious; it seems unlikely to be tolerance of WF histocompatibility antigens per se, since a few tolerant rats did become diabetic and their disease was capable of destroying transplanted WF islet tissue, despite the fact that transplanted WF skin was accepted. Another possibility is destruction by the WF cells of a "forbidden clone" of BB cells responsible for autoimmune diabetes in unmodified BB rats. Perturbations in the immunological network may be the underlying cause of diabetes if autoimmunity is involved. In this regard there is experimental evidence that T cell immunoregulatory malfunction is a dominant factor in the etiology of autoimmune disease (*14*). Persistence in BB rats of a population of lymphohematopoietic cells from normal nondiabetes-prone WF donors could play a role in keeping an autoimmune process in abeyance. The chimeric cells might, for example, correct possible deficiencies in BB lymphocyte subpopulations—such as suppressor cells. Alternatively, if the syndrome is triggered by a yet to be detected virus, the chimeric cells could reequip the BB hosts with competent antiviral clones which they may lack. Study of these possible mechanisms in this animal model should provide fruitful avenues for future investigations of etiology and therapy of diabetes.

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8. Two of these recipients that suffered recurrent diabetes were challenged with a second WF skin graft which they permanently (more than 100 days) accepted, again confirming the persistence of the tolerant state.
9. All of these rats remained normoglycemic through a 1- to 4-month observation period after which they were killed and found histologically to have healthy transplanted WF islets.
10. Statistical analysis was performed by a chi-square test.
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Retrograde Amnesia: Possible Role of Mesencephalic Reticular Activation in Long-Term Memory

Abstract. *A patient with cerebral trauma recovered considerably from the resulting anterograde but not retrograde amnesia. The persistence of retrograde amnesia is attributed to a lesion in the ventral tegmental region, which suggests a role for mesencephalic reticular activation in long-term retrieval.*

Two types of memory deficits are commonly identified: anterograde amnesia (AA), an inability to learn and retain new information after the onset of pathology; and retrograde amnesia (RA), an inability to retrieve information acquired prior to the onset of pathology, usually involving loss of remote memory (*1*). Parallel recovery from AA and RA is a common clinical finding (*2*), as is RA recovering first with AA recovering later or not at all (*3*).

Double dissociation between the two types of amnesia and the presence of RA without AA have been described (*1, 4*), but cases are rare and sparsely documented. We now report a case of considerable recovery from AA but not RA, which may offer insights into mechanisms of long-term retrieval.

A 36-year-old, right-handed, college-educated male had an open skull fracture in the right parieto-occipital and temporal areas and herniation of the right hemisphere with compression of the left mesencephalon at the tentorial notch. In surgery, a small portion of macerated brain was removed, the dural laceration was sewn over, and the bone-plate replaced. Six weeks later, a ventriculoatrial shunt was performed. There was hemiparesis with spasticity of the distal upper right extremity, a right Babinski's sign, and a left superior quadrantanopsia. The patient was disoriented, his speech anomic and agrammatic. There was profound AA: his recall of seeing a person did not exceed 2 to 3 minutes, and his ability to retain names was still shorter. There was also profound RA. The patient maintained that he was 16 to 18 years old and mentioned his parents' address as his residence. He revealed no knowledge of his subsequent life history, his marriage, children, or past employment. His command of general information was equally impaired.

During the 2-year course of recovery the patient became fully oriented; linguistic and motor deficits virtually disappeared, but the quadrantanopsia remained. Recent memory was improving. A continuity of experience became possible; he could keep track of weekly and monthly events and retain information from newspapers and television. There was no parallel recovery from the 20-year deep RA. The patient's past history was reconstructed for him, but lacked a sense of authenticity. Command of general information did not improve beyond what he had been taught during this time; he could not answer questions like, "Who wrote *Hamlet*?" or "What is the capital of France?"

Table 1 summarizes the patient's per-

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formance on tests of memory over a 1.5-year period. Tasks of recent memory—the Wechsler Memory Scale (*5*), and the Buschke Selective Reminding Test (*6*)—showed significant improvement, while tests of remote memory—the Boston Retrograde Amnesia Battery (*7*), and the general knowledge battery (*8*)—did not (except the "recall" part of BRA, where the scores were lowest). The Wechsler memory quotient changed from 86 to 106, and the Wechsler Adult Intelligence Scale IQ from 86 to 105 (verbal, 90 to 112; performance, 82 to 95). Both memory and IQ are now within the normal range and mutually consistent (*9*). Performance on the other tests remains deficient.

The patient's performance on verbal recognition subsections of the general knowledge battery was inversely related to the degree of categorical proximity between correct and alternate choices. Pairwise comparisons of adjacent steps were significant (*10*) (Closest, 43 percent correct; intermediate, 58 percent; $z = 1.70$, $P < .05$. Intermediate, 58 percent; distant, 77 percent; $z = 2.47$, $P < .01$). Such dissociation has been interpreted as related to retrieval rather than to storage (*11*).

A series of computerized tomographic (CT) examinations was performed during the early course of recovery (in 1977 and 1978), through the use of sections 8 mm thick and supplemental coronal and overlapping sections. Findings included moderate ventricular enlargement, a region of rarefaction in the right middle and posterior temporal areas, and a small region of rarefaction along the left mid-temporal convexity. The ventricular shunt tip was in place within the lateral ventricle. The findings were consistent with atrophic changes within the temporal lobes, as a result of both trauma and surgery.

Ventricular dilation (thus probable increased intracranial pressure, periventricular atrophy, or both) and bilateral temporal damage (both convexital and mesial) have been implicated in memory deficits, but not with RA as the predominant and relatively isolated component (*12*). It was concluded that the CT findings could not fully account for our patient's condition.

Since relative recovery from AA in the presence of persistent severe RA has not been interpreted neuroanatomically, some speculation was necessary. Reverberation in cortico-thalamic-limbic loops may constitute a crucial stage in consolidation (*13*). It is possible that long-term retrieval involves similar stages, but in a reverse order. It could be hypothesized further that, whereas in the process of consolidation the reverberation can be initiated by external stimulation, for retrieval some intrinsic reticular activation is necessary. These considerations led to the decision to repeat CT studies (in May 1979) with particular attention to the mesencephalic tegmentum, and with sections 5 mm thick.

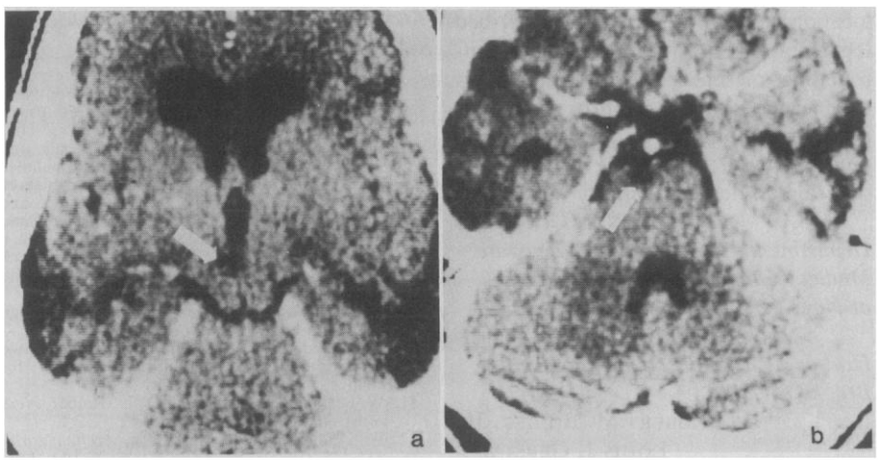

Fig. 1. Computerized tomographic scan images of the mesencephalic lesion. Transverse sections, each 5 mm thick, demonstrating the lesion extending from (a) the ventral tegmental portion of the upper mesencephalon to (b) the ventral portion of the pontomesencephalic junction. The patient's left is to the reader's left.

In addition to confirming the earlier findings, these studies revealed a narrow band of hypodensity in the median and left paramedian zones, extending from the ventral tegmental portion of the upper mesencephalon (where the midline was reached and crossed) caudally to the ventral portion of the ponto-mesencephalic junction (Fig. 1). This was confirmed by coronal reconstruction.

Table 1. Performances on standardized tests at beginning (T_1) and end (T_2) of a 1.5-year interval. Scores are expressed as percentages of maximum possible scores. One-tailed tests of difference between two proportions with correction for chance in the case of multiple-choice tasks were used.

Task	T_1	T_2	z
Wechsler Memory			
Total score	53.2	69.4	2.18*
Buschke Selective Reminding			
Total recall	53.1	91.5	4.67†
Long-term storage	36.1	80.0	5.55†
Long-term retrieval	20.0	74.6	7.51†
Consistent retrieval	0	56.2	9.25†
Boston Retrograde Amnesia			
Recognition	36.7	43.3	0.75
Recall	11.7	30.0	2.47*
Famous faces	27.4	39.7	1.58
General knowledge battery			
Verbal recall	58.3	66.7	0.84
Verbal recognition	64.8	59.3	−0.69
Visual recognition	30.8	28.5	−0.18

*$P < .02$. †$P < .0001$.

Further analysis revealed changes in average density within the region where the ventral tegmental nucleus is found and a component of the medial forebrain bundle originates. In its caudal extension, the hypodense region probably overlapped with the trajectory of locus coeruleus projections into the medial forebrain bundle. There were no abnormalities in dorsomedial or anterior thalamic nuclei, mammillary bodies, or the temporal stem.

The ventral tegmental area is identified with the ventral tegmental pathway—a subdivision of the cholinergic reticular formation projecting into limbic structures (*14*). It constitutes a major source of the ascending portion of the medial forebrain bundle, which in turn projects into the hippocampi via the medial septum (*14*) and into the mammillary bodies via the mammillary peduncle (*15*). The locus coeruleus projections into the medial forebrain bundle are the major sources of noradrenergic influences on the septum, hippocampi, and other limbic structures (*16*).

In our patient, ascending reticular projections into limbic structures most often implicated in memory (hippocampi and mammillary bodies) seem to have been severed, while those into the thalamus and neocortex were spared. That this was the apparent cause of profound RA in the absence of comparably severe AA or a general arousal deficit may indicate that selective mesencephalic reticular activation of limbic structures constitutes a fundamental component of long-term retrieval. The importance of such activation has been demonstrated in animals (*17*). The rarity of this pattern of memory deficits can be attributed to the fact that only a small lesion of precise

location can affect the above described tegmental area without affecting surrounding mesencephalic structures. If the mesencephalic structures are affected, the resulting impairment to the patient's overall arousal level (*18*) will override a selective memory deficit.

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Characterization of a 41-Residue Ovine Hypothalamic Peptide That Stimulates Secretion of Corticotropin and β-Endorphin

Abstract. *A peptide with high potency and intrinsic activity for stimulating the secretion of corticotropin-like and β-endorphin–like immunoactivities by cultured anterior pituitary cells has been purified from ovine hypothalamic extracts. The primary structure of this 41-residue corticotropin- and β-endorphin–releasing factor has been determined to be:*

H-Ser-Gln-Glu-Pro-Pro-Ile-Ser-Leu-Asp-Leu-Thr-Phe-His-Leu-Leu-Arg-Glu-Val-Leu-Glu-Met-Thr-Lys-Ala-Asp-Glu-Leu-Ala-Gln-Gln-Ala-His-Ser-Asn-Arg-Lys-Leu-Leu-Asp-Ile-Ala-NH_2

The synthetic peptide is active in vitro and in vivo.

Experimental and clinical observations have supported the concept that the hypothalamus plays a key role in the regulation of the secretory functions of adenohypophyseal corticotropic cells (*1*). More than 25 years ago, Guillemin and Rosenberg and Saffran and Schally independently demonstrated the presence of factors in hypothalamus that would increase the rate of corticotropin secretion by the pituitary gland incubated in vitro or maintained in organ culture (*2*). Several characterized substances present in hypothalamic extracts release corticotropin in vitro or in vivo including vasopressins, norepinephrine, fragments of proteins such as hemoglobin or myelin basic protein, and modified amino acids (*3, 4*). None of the previously characterized secretagogs, with the possible exception of norepinephrine which has yet to be fully evaluated, meets the criteria (*5*) expected of a physiologic corticotropin-releasing factor (CRF). We report here the purification, sequence analysis, and total synthesis of a 41-residue peptide that stimulates the secretion of corticotropin-like and β-endorphin–like immunoactivities in vitro and in vivo.

Throughout our purification program we used an in vitro method for assaying the ability of a putative CRF to stimulate the secretion of corticotropin (ACTH) by primary cultures of rat pituitary cells (*6*). Concentrations of ACTH and β-endorphin (β-End) in culture fluids were assayed by double antibody radioimmunoassays (RIA's) (*7*). Although the terms ACTH and β-End are used in this report, it is recognized that these RIA's detect multiple forms of the peptides; for example, the β-endorphin RIA measures pro-opiocortin, β-lipotropin, and β-endorphin and its acylated forms.

Starting material for this purification was a side fraction of 490,000 fragments of ovine hypothalamus (initially processed in the Laboratories for Neuroendocrinology at the Salk Institute) during the program to characterize gonadotropin-releasing hormone. As described (*8*), tissues were extracted in a mixture of ethanol, acetic acid, and chloroform, defatted with a mixture of ether and petroleum ether and partitioned in the system consisting of 0.1 percent acetic acid, *n*-butanol, and pyridine (11:5:3). Although ACTH-releasing activity was found in portions of both upper and lower phases, only the lower phases were available to us for further purification and thus were used as starting material for the project we describe.

After ultrafiltration (Amicon UM-10) or dialysis (Spectrapor 3) against 2*N* acetic acid, about 350,000 fragment equivalents of the retained fractions (weighing 15 g) were subjected to gel filtration on Sephadex G-50. The bulk of material was chromatographed at 4°C (*9*) in successive runs on a G-50 column, 3.1 by 150 cm, eluted with 2*N* acetic acid. Two zones of ACTH-releasing activity were detected: zone 1 eluting at about 1.3 V_e/V_o and zone 2 eluting at about 2.0 V_e/V_o. Multiple ACTH-releasing zones, including "large CRF's," have been described (*4, 10*). The two zones showed

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