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6. Care was taken with infiltration anesthesia to minimize discomfort. Successful conditioning performance demands the animals be in no distress, since discomfort is invariably reflected in high baseline heart rates and a lack of conditioning.
7. The heart rate response was measured by subtracting the number of beats in the 6-second period immediately preceding the light (baseline period) from the number of beats during the light period. This difference was expressed in beats per minute (*3*).
8. The rationale for separating the stimulus period into a 500-msec (phasic) period and a subsequent 5500-msec tonic period for analytic purposes is that neurophysiological analyses of other segments of the system have indicated distinct transient responses occurring within the first few hundred milliseconds (*9*). This is particularly prominent for the cardiac sympathetic postganglionic neurons, in which the light-evoked response is almost entirely phasic. In fact, the CR may be largely mediated by this short-latency component (*2*).
9. C. M. Gibbs and D. H. Cohen, *Soc. Neurosci. Abstr.* **6**, 424 (1980); J. Wall *et al.*, *ibid.*, p. 424.
10. Supported by NSF grants BNS-72-20537 and BNS-80-16396 and NIH grant P01 NS 14620. M.R.G. was supported by NIH training grant T32 HL07284.. We thank C. Schack for her secretarial assistance.
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Supraoptic Nucleus of the Brattleboro Rat Has an Altered Afferent Noradrenergic Input

Abstract. *The distribution of fluorescent varicosities in the supraoptic nucleus of Brattleboro rats was compared to that in normal rats. The Brattleboro rat, which is characterized by a genetic absence of vasopressin, had fewer fluorescent varicosities in apposition to the vasopressin-deficient perikarya. The oxytocin-producing neurons in the same nucleus were hyperinnervated. These data suggest that the target neuron peptide (vasopressin) is necessary for the maintenance of normal noradrenergic innervation patterns.*

Oxytocin and vasopressin are magnocellular neurosecretory peptides of the mammalian hypothalamo-neurohypophyseal system. They play an important role in the onset of parturition and milk ejection and help regulate water and electrolyte balance within intra- and extracellular fluid compartments. These hormones are synthesized in and transported by neurons of the paraventricular nucleus (PVN) and supraoptic nucleus (SON) of the hypothalamus (*1*). In addition, the hypothalamic nuclei receive a dense input from brainstem noradrenergic neurons of the locus coeruleus and pontomedullary reticular formation (*2*). These noradrenergic neurons, through their hypothalamic innervation, are thought to play an important role in regulating the release of vasopressin and oxytocin (*3*).

A new approach for examining putative neuronal interactions (*4*) has been used to study the distribution of noradrenergic varicosities and their peptidergic target neurons in the SON and PVN (*5*). From these studies it appears that the densest accumulation of fluorescent varicosities occurs in the most ventral regions of the SON, where vasopressinergic cells predominate. On the basis of a recent ontogenetic study wherein peptide staining of neurosecretory neurons of the SON and PVN appeared before fluorescence of noradrenergic varicosities, it was postulated that the magnocellular neurosecretory products act as neurotrophic agents, providing a target for ingrowing noradrenergic axons (*6*). Examining a different central target, however, Lauder and Bloom (*7*) found, through the use of autoradiography and fluorescence histochemistry, that noradrenergic neurons of the locus coeruleus begin their growth and development several days before their probable target neurons in the cerebellum and hippocampus begin to differentiate. This suggested that monoamine neurons regulate the onset of neuronal differentiation in their projection areas, perhaps in a neurotrophic manner.

To further test these two hypotheses, we studied the Brattleboro rat, which lacks a specific target neuron peptide (vasopressin) but not the target neuron itself. This rat is genetically incapable of vasopressin synthesis and is therefore chronically dehydrated (*8*). Analysis of noradrenergic innervation patterns of the vasopressin-deficient SON might help determine whether peptide content is necessary for the maintenance of functional noradrenergic interactions.

Six adult male Long-Evans homozygous Brattleboro rats and five normal male Long-Evans rats of the same age were decapitated and processed for formaldehyde-induced fluorescence of catecholamines (*9*). All tissue blocks were serially sectioned (6 μm) and every tenth section was mounted and stained with Luxol fast blue and cresyl violet to provide anatomical orientation. Sections throughout the rostral, middle, and caudal levels of the SON were pressed onto glass slides and photographed in a Leitz Dialux fluorescence microscope with mercury-vapor lamp, narrow-band excitation, and K460 barrier filters. Sections adjacent to those used for fluorescence microscopy were mounted on gelatin-coated glass slides and stained for rat neurophysin by the peroxidase-antiperoxidase procedure (*10*). The sections stained for neurophysin were photographed, and photomontages were assembled to reconstruct the SON at its various levels. Using clear acetate overlays, we traced all the neurons of the SON, marking those which stained for neurophysin and, in the Brattleboro rat, marking those which did not stain for neurophysin. Each overlay was then transferred to the photomontage of the adjacent section processed for fluorescence microscopy, and the varicosities abutting both neurophysin-positive and neurophysin-negative perikarya were marked. With this approach the pattern of fluorescent varicosities in the SON of three normal and three Brattleboro rats was quantitatively assessed. We counted the number of varicosities apposing each of a total of 1415 neurons (Table 1).

Table 1. Number of varicosities apposed to perikarya in the SON of normal and Brattleboro rats. Each value is the mean for three rats (*15*).

SON region	Strain	Varicosities per neuron	*P**
Dorsal	Control	1.25	
	Brattleboro	1.76	< .0005
Ventral	Control	1.97	
	Brattleboro	1.64	< .001

*One-way analysis of variance.

The SON of control animals contained a dense distribution of catecholamine varicosities. The distribution was densest in the ventral region of the nucleus and became progressively less dense in the more dorsal regions (Fig. 1). Most of the ventrally distributed varicosities appeared to appose magnocellular perikarya and their ventrally directed processes. The majority of these neurons have been immunocytochemically identified as vasopressinergic, whereas most of the dorsal SON neurons are oxytocin-containing (*11*).

The distribution of noradrenergic fibers in the SON of the Brattleboro rats appeared different from that in the controls. Ventral regions of the nucleus

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were characterized by neurons devoid of neurophysin and vasopressin (Fig. 2) and by a paucity of catecholamine varicosities (Fig. 3), which were observed only occasionally in apposition to vasopressin-deficient neurons. In control animals neurons in this same region of the nucleus were often bordered by as many as seven varicosities (as seen in a single tissue section). Moreover, the dorsally situated oxytocin neurons appeared to be more heavily contacted by varicosities in the Brattleboro rats than in the controls.

The apparent difference between strains in the distribution of noradrenergic fiber terminals in the SON was verified by the quantitative procedure described earlier. There were 12 percent fewer varicosities apposing vasopressin-deficient perikarya and 14 percent more varicosities apposing oxytocin-producing perikarya (Table 1) in the Brattleboro rats compared to the controls. These differences confirm the qualitative assessment and are statistically significant.

In summary, we found that the noradrenergic innervation of peptide-depleted neurons in vasopressin-deficient regions of the Brattleboro rat SON is abnormal and that, in the dorsal region of the nucleus, the perikarya of oxytocin-producing neurons were contacted more frequently by fluorescent varicosities than in controls. Although neuronal plasticity has been observed in response to injury, the present study indicates a similar phenomenon resulting from chronic peptide depletion in target neurons.

Although the quantitative data collected for this study verify our morphological observations, the differences between the experimental and control animals were not as great as the visual observations implied. This discrepancy may be explained in part by the larger size of individual neurons in the SON of Brattleboro rats producing an overall appearance of decreased fluorescence. These data, however, suggest that vasopressin is essential for normal noradrenergic input to the SON.

The magnocellular neurons in the SON may contain other hormones than vasopressin and oxytocin. For example, peptides of the gastrin family frequently coexist with oxytocin in the neurons of the SON and PVN (*12*). Also, the opiate peptide dynorphin is present in the vasopressin-deficient neurons of the Brattleboro rat (*13*). Since it is unknown whether norepinephrine plays a role in synthesizing or secreting these hormones, noradrenergic input may be at least partly responsible for controlling their function. This could explain the incomplete absence of fluorescent varicosities in the Brattleboro rat (*14*).

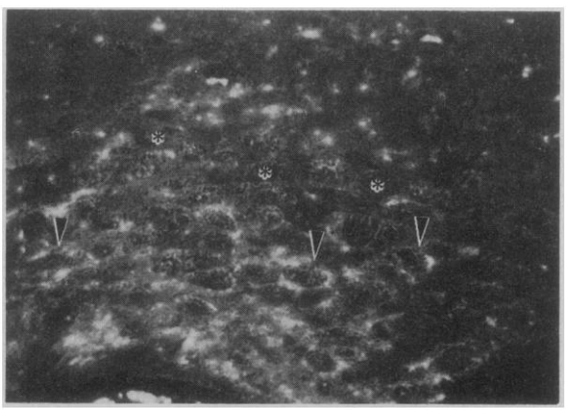

Fig. 1. Coronal section through the SON of a normal Long-Evans rat following histofluorescence preparation. The fluorescent catecholaminergic varicosities abut magnocellular neurons, especially in the ventral regions of the SON (arrowheads), while more dorsally the neurons are contacted less frequently (asterisks) (×120).

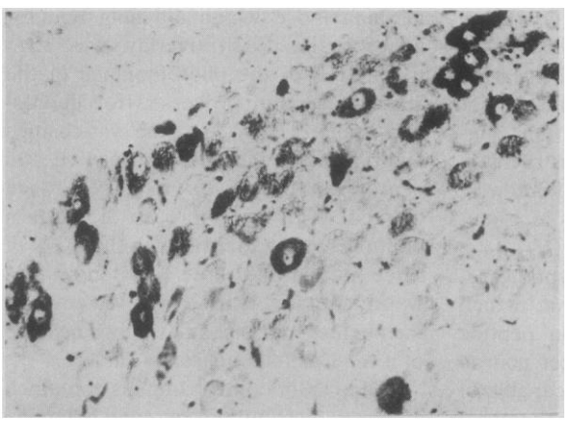

Fig. 2. Coronal section through the SON of the Brattleboro rat following neurophysine immunocytochemistry and counterstaining with cresyl violet. Oxytocin-containing neurons are present in the dorsal portion of the nucleus, while the ventral portion contains neurons deficient in neurophysin and vasopressin (×135).

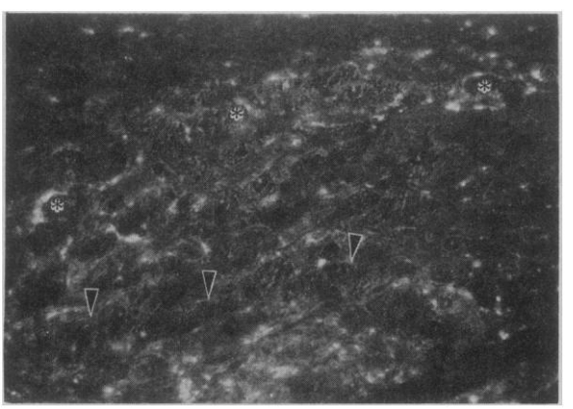

Fig. 3. Coronal section through the SON of the Brattleboro rat following histofluorescence preparation. In addition to the larger size of individual neurons compared to those in controls, there is less innervation by catecholamine cells ventrally (arrowheads) and a greater number of varicosities in the dorsal, oxytocin-rich portion (asterisks) (×120).

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14. In addition to developmental studies to determine whether normal contacts existed initially, it is essential to perform studies to determine whether the lack of noradrenergic varicosities is the result of a peptide deficiency or merely the result of an increased turnover rate in the terminals as a result of chronic dehydration.
15. These data were determined by dividing the SON into dorsal and ventral regions representing oxytocin- and vasopressin-rich regions, respectively, as determined with immunohistochemical techniques in normal rats and verified with neurophysin staining in the homozygous Brattleboro rat (*11*).
16. We thank J. Fields for skilled technical assistance. Antiserum to rat neurophysin was provided by A. G. Robinson through PHS grant AM 16166. This work was supported by PHS grants NS 15816, AG 00847, and MH 14577.
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Oculomotor Reaction Time in Dementia Reflects Degree of Cerebral Dysfunction

Abstract. *The effects of diffuse cerebral dysfunction on oculomotor reaction time were assessed in patients with dementia of presumed Alzheimer's etiology and in normal age-matched control subjects. Patients were classified into mild, moderate, and severe groups on the basis of independent neurological, neuropsychological, and neuroradiological ratings for disease severity. Saccadic latencies to targets appearing in parafoveal and near peripheral vision showed significant increases from the normal controls to dementia groups, with each severity subdivision clearly differentiated from the others in terms of mean oculomotor reaction time. These data offer strong evidence for a direct relationship between degree of cortical structural integrity and simple oculomotor reaction time and suggest a higher cortical regulatory role in sensory-motor integration.*

For more than 130 years, reaction time (RT) has been examined experimentally as a measure of conduction speed of the central nervous system. As early as 1850, Helmholtz (*1*) used an RT measure to estimate the speed of transmission along a frog's motor nerve. Reaction time has been used as an index of cerebral dysfunction in a variety of brain-damaged groups, with latencies exceeding normal values (*2–5*). Prolonged RT's on both the ipsilateral and contralateral side in patients with unilateral cerebral disease (*4*) suggest that RT may reflect the degree of overall cerebral intactness rather than specify localized damage. Correlations of RT to severity of brain injury or dysfunction (*5*) argue further for an association between this simple sensory-motor task and the extent of involvement of the central nervous system's cortical substrate. Our study examined the relationship between RT performance and the severity of dementia (and by inference, cerebral integrity) in patients with presumed dementia of the Alzheimer's type (DAT) (*6*).

Such dementia (*7*) is marked by progressive cerebral degeneration of unknown etiology, producing cognitive and psychomotor disturbances. Alzheimer's is believed to be the most frequent cause of dementia (*8*). Morphologically, DAT is characterized by ventricular dilation and cortical atrophy most prominent in the frontal and temporal lobes. Microscopically, the degenerative changes include senile plaque formation, neurofibrillary tangling, and granulovacuolar degeneration.

Simple oculomotor RT was chosen as the experimental measure because this task minimizes attentional and vigilance factors, since short saccades to stimuli have been traditionally considered volitional but highly automated (*9*). Oculomotor RT of 12 normal age- and sex-matched elderly control subjects and 12 patients with DAT were obtained (*10*). The DAT patients were further divided into mild, moderate, and severe groups (four per group) on the basis of independent neurological, neuropsychological, and neuroradiological ratings for disease severity. A deterioration index derived from these separate evaluation procedures consisted of the average ranking on these severity scales (from 1, minimum impairment, to 5, severe impairment) (*11*).

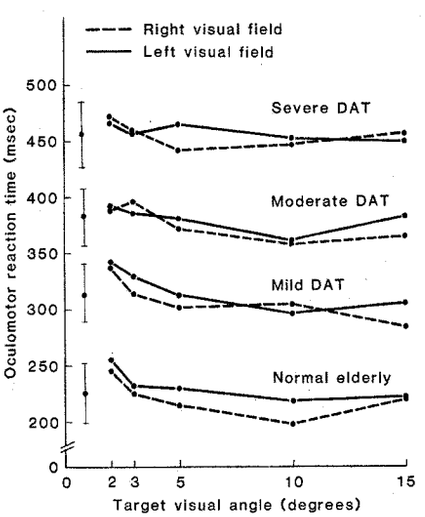

Fig. 1. Group mean oculomotor RT's as a function of visual angle and visual half-field. Means (± standard deviations) across target visual angles and visual fields are shown at the left.

Saccadic latencies were obtained with an eye-movement recorder (Biometric model 200) paired with a digital processor (Nova 12/20). Three-letter consonant-vowel-consonants (CVC's) displayed on a cathode-ray tube (CRT) (Lexiscope) served as the stimuli. The CVC's in the right visual field had the first letter beginning at a visual angle of 2°, 3°, 5°, 10°, or 15°, with the last letter of the CVC ending at similar angles in the left field trials. All stimuli subtended 1° of horizontal visual angle and were displayed for 1 second. A short practice and calibration session for the photoelectric sensors preceded the actual experimental session. Eight trials at each of the ten positions presented in a random sequence (sampling without replacement) constituted the experimental session. Each subject rested his head in a chin and forehead rest with the glabella 40 cm away from the CRT. We initiated trials with a "ready" command when subjects were fixated on the zero point on the CRT. Subjects were instructed to move their eyes to the location of the stimulus and name the word. Oculomotor RT was defined as the interval between stimulus onset and time of movement to the appropriate half-field of 0.5°, an experimentally determined value used to separate target saccadic movements from ongoing oscillatory eye jitter. Latency windows of 100 to 400 msec for normal subjects and 100 to 1000 msec for patients were used to accept saccadic RT values; approximately 4 percent of normal subjects' and 8 percent of DAT patients' trials fell outside these limits and were not analyzed. Eye position was sampled at 1000 points per second for the 1-second period after stimulus onset.

Data analyses were based on subject median RT values, as some tendency to positive skewing was evident in the patient group. The overall latency of the DAT patients was 158 msec longer than that of the normal aged controls (Fig. 1). Analyses of variance (*12*) demonstrated that DAT patients displayed significantly ($P < .001$) longer saccadic latencies than normal subjects and that the mild, moderate, and severe groups in turn had significantly ($P < .001$) different latencies from one another. Group separation

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