Cardiovascular Input to Hypothalamic Neurosecretory Neurons

L. P. RENAUD,¹ J. H. JHAMANDAS, R. BUIJS,²
W. RABY AND J. C. R. RANDLE

*Neurosciences Unit, Montreal General Hospital and McGill University
Montreal, Quebec Canada H3G 1A4*

RENAUD, L. P., J. H. JHAMANDAS, R. BUIJS, W. RABY AND J. C. R. RANDLE. *Cardiovascular input to hypothalamic neurosecretory neurons*. BRAIN RES BULL 20(6) 771-777, 1988.—In vivo extracellular recordings from rat supraoptic and paraventricular magnocellular neurosecretory cells (MNCs) indicate that putative vasopressin-secreting MNCs may be identified by an abrupt and brief cessation in firing consequent to a transient drug-induced rise in arterial pressure sufficient to activate arterial baroreceptors. In the diagonal band of Broca (DBB), a population of neurons projecting towards the supraoptic nucleus are activated during this drug-induced hypertension. Electrical stimulation in DBB selectively depresses supraoptic vasopressin-secreting MNCs. Intracellular recordings in perfused hypothalamic explants confirm a DBB-evoked bicuculline-sensitive and chloride-dependent postsynaptic inhibition, similar to that associated with the application of γ -aminobutyric acid (GABA) in approximately half of supraoptic MNCs. Since bicuculline also selectively blocks baroreceptor-induced inhibition in supraoptic MNCs, it is proposed that the depressant baroreflex input to vasopressin-secreting MNCs involves a population of DBB neurons and GABAergic interneurons located close to MNCs. An excitatory and selective input to vasopressin-secreting MNCs follows chemoreceptor activation, possibly mediated by the A1 noradrenergic cell group in the ventrolateral medulla. Another excitatory input to both vasopressin- and oxytocin-secreting MNCs is triggered by circulating angiotensin II and appears to be relayed centrally through an angiotensinergic projection from the subfornical organ.

Supraoptic nucleus	Neurosecretory cells	Baroreceptors	Chemoreceptors	Gamma-aminobutyrate
Norepinephrine	Electrophysiology	In vivo recordings	In vitro recordings	

THE magnocellular neurosecretory cells (MNCs) of the hypothalamic supraoptic (SON), paraventricular (PVN) and accessory magnocellular nuclei function to differentially regulate circulating levels of oxytocin and vasopressin. These hormones are synthesized in separate populations of MNCs [45,51]. Their release from storage vesicles in the neural lobe occurs in proportion to the frequency and patterning of electrical activity in the neurohypophyseal axis [4, 8, 12, 15]. One approach towards understanding the central mechanisms involved in regulating this neurosecretory process has focused on the neurophysiology of MNCs themselves, notably their intrinsic membrane properties and synaptic inputs [6, 35, 39-41]. The latter are considered further in this presentation which extends earlier electrophysiological observations on the responses of rat MNCs to inputs of cardiovascular origin [53].

VASOPRESSIN VS. OXYTOCIN-SECRETING MNCs

Extracellular recordings from antidromically identified MNCs in anesthetized rats generally reveal two categories of

neurons defined on the basis of firing patterns and sensitivity to a brief rise in arterial pressure sufficient to activate peripheral baroreceptors. Putative oxytocin-secreting MNCs (Fig. 1) display an irregular slow continuous firing pattern that does not respond to baroreceptor activation. Recent studies have further indicated that this type of MNC is selectively excited by systemic cholecystikinin and by gastric distention [42]. In lactating females such MNCs discharge prior to milk ejection [31,35]. Putative vasopressin-secreting MNCs (Fig. 2) typically demonstrate either a phasic or continuous firing pattern, an abrupt but transient cessation in activity consequent to a drug-induced rise in arterial pressure and can be prompted to initiate a discharge by chemoreceptor stimulation [16,17].

BARORECEPTOR INPUT TO MNCs

Although peripheral baroreceptor stimulation affects neuronal excitability in various levels of the neural axis [19, 25-28, 30, 33, 34, 48], there is considerable uncertainty as to the pathways whereby this information ascends beyond the

¹Requests for reprints should be addressed to Dr. Leo Renaud, Neurosciences Unit, Montreal General Hospital, 1650 Cedar Avenue, Montreal, Quebec Canada H3G 1A4.

²Permanent address: Netherlands Institute for Brain Research, Meibergdreef 33, 1105 AZ, Amsterdam, The Netherlands.

FIG. 1. Several features deemed typical of extracellular recordings from putative oxytocin-secreting MNCs in the SON of the rat. Top pair of records: data (from [42]) illustrate mean arterial pressure (MAP) and simultaneous ratemeter records from a continuously firing neuron. Firing frequency does not change during a brief rise in MAP induced by administration of metaraminol (Met, 4 µg IV at arrow) but increases significantly during distention (5 cc) of an intragastric balloon. Middle pair of records: data (from [42]) illustrate that intravenous administration of cholecystikinin (CCK-8) provokes a prolonged increase in firing from another blood pressure-insensitive, continuously active MNC. Lower records: Data (from [31]) in a lactating suckled rat display a slow irregular background firing, a brief burst of high frequency firing (top integrated trace, lowest unit trace) and subsequent rise in intramammary pressure (centre trace) which results in milk ejection.

nucleus of the solitary tract [2, 17, 21]. Recently, interest has been directed towards the diagonal band of Broca (DBB) where a population of cells demonstrate antidromic activation from the area of SON and also increase their activity during a drug-induced rise in blood pressure (Fig. 3). Since electrical stimulation within DBB, which contains an abundance of GABA-synthesizing neurons [7], selectively depresses the firing of vasopressin-secreting MNCs [20,21], one hypothesis is that the DBB contains a population of GABAergic inhibitory neurons which are activated during baroreceptor stimulation and which in turn project towards the SON (and PVN) to suppress the firing of vasopressin-secreting MNCs [21].

Characterization of the baroreceptor-evoked depression of vasopressin-secreting MNCs has been possible through several neuropharmacological sequences. Earlier proposals

FIG. 2. Several features deemed typical of extracellular recordings from putative vasopressin-secreting MNCs in the SON of the rat. Top pair of traces: lower ratemeter records [9] of a phasically-active MNC illustrate premature cessation of a burst during a metaraminol-induced rise in MAP. Middle pair of traces: (also from [9]) similar sensitivity of a continuously-firing MNC during a metaraminol-induced rise in MAP. Lower records: Data (from [17]) illustrate the induction of a phasic burst of ipsilateral (i) but not contralateral (c) chemoreceptor activation achieved by instillation of carbon dioxide saturated saline into the carotid sinus.

that noradrenergic fibers might mediate baroreceptor-evoked inhibition of MNCs have been discounted by recent data on the alpha-mediated excitatory actions of noradrenaline and noradrenergic inputs of MNCs [9-11, 37, 52], and by persistence of baroreceptor-evoked depressions following 6-hydroxydopamine-induced depletion of catecholamines in SON and PVN [9,11]. However, local pressure applications of a GABA-A receptor antagonist bicuculline methiodide (100 µM) selectively and reversibly abolish the depression of SON [23] and PVN [29] vasopressin-secreting MNCs consequent to baroreceptor activation (Fig. 4). In view of the failure of other known receptor antagonists to induce a similar effect, these data support the hypothesis stated in the preceding paragraph, i.e., that GABA mediates the baroreceptor-activated inhibitory input to vasopressin-secreting MNCs. In view of the reciprocal patterns of firing among certain DBB neurons and SON MNCs when arterial pressure is transiently elevated (Fig. 3), and the ability for bicuculline to reversibly and selectively block DBB-evoked

FIG. 3. Intravenous metaraminol ($2-4 \mu\text{g}$, arrow) produces a transient rise in mean arterial pressure (top trace). During this response, certain central neurons may reveal reciprocal activity patterns. In the middle ratemeter record (from [19]), a silent diagonal band of Broca (DB) neuron (antidromically activated from an electrode in the SON area) demonstrates an increase in activity. The lower trace from a phasically-active SON MNC (recorded independently) reveals a transient cessation in on-going activity.

FIG. 4. Data (from [23]) illustrate the ability for pressure applied bicuculline (A), a GABA antagonist, but not prazosin (B), an α_1 adrenoreceptor antagonist, or timolol (C), a β adrenoreceptor antagonist, to reversibly block the anticipated reduction in firing of three different phasically-active SON MNCs consequent to a metaraminol-induced (arrowhead) rise in blood pressure.

inhibition of SON vasopressin-secreting MNCs (Fig. 5), it is reasonable to propose involvement of DBB neurons in this central baroreflex activity.

A more detailed electrophysiological and pharmacological characterization of the DBB input to SON MNCs has ensued from intracellular recordings from SON neurons in

vitro in perfused hypothalamic explants [36]. Recordings with potassium acetate-filled micropipettes reveal an abundance of spontaneous inhibitory postsynaptic potentials (IPSPs) which peak in 3–5 msec and decay exponentially with a mean time constant of 20.2 msec, i.e., 1.5-fold greater than the membrane time constant of 13.8 msec ($n=9$ cells).

FIG. 5. Poststimulus histograms from a phasic SON MNC in response to stimulation in the diagonal band (DB) and posterior pituitary (PP, to monitor the antidromic spike). The upper control trace illustrates a brief decrease in firing induced by the DB stimulus. In the presence of a local pressure-applied solution of 100 μ M bicuculline (middle trace) this depression is abolished, but returns shortly after the application has terminated (lowest trace). Similar data are presented in [20].

In approximately 50% of SON neurons, DBB stimulation evokes a compound IPSP with a mean latency of 6.8 msec. These IPSPs peak in 3–10 msec and decay exponentially over 60–100 msec with an IPSP time constant of decay of 37.0 msec ($n=16$ cells), approximately 2.5 times their mean cell time constant. Membrane conductances increase from 0.17 to 3.0 nS during spontaneous IPSPs, and from 0.8 to 22.0 nS during evoked IPSPs. The following observations indicate that these conductances reflect an increase in membrane chloride permeability. IPSP reversal potentials (approximately -80 mV) shift in a more depolarized direction when chloride ions are injected intracellularly or lowered in the extracellular media. IPSPs are also markedly prolonged by pentobarbital and abolished in the presence of bicuculline, further features of a GABA-mediated postsynaptic inhibition. Based on the similarities between these observations and extracellular data obtained in vivo, it is further suggested that this in vitro input is analogous to the in vivo baroreceptor-activated inhibition of SON vasopressin-secreting MNCs.

However, anatomical data do not generally support the existence of a direct (i.e., monosynaptic) GABAergic path-

FIG. 6. Data (from [36]) in A illustrate the DB-evoked compound inhibitory postsynaptic potentials (IPSPs) recorded in SON MNCs. These are positioned after the onset of an intracellular current pulse so as to illustrate the IPSP reversal. The voltage-current plots in B demonstrate a prominent increase in membrane conductance at the peak of the IPSP.

way from DBB to MNCs. Anterograde and retrograde tracer studies [24, 44, 46, 49] indicate that DBB neurons project mainly to the perinuclear zone around the SON and PVN. Given that this area also contains GABAergic neurons [18,47], it would appear that the DBB-evoked IPSPs in SON MNCs are relayed disynaptically via the perinuclear zone. Hence, perinuclear neurons may be the source for the heavy GABAergic innervation noted on MNCs [47,50] and mediate both the DBB and baroreceptor-evoked inhibition of vasopressin-secreting MNCs.

Earlier reports of GABA's actions on rat MNCs based on extracellular observations [1, 5, 35, 41] have now been supplemented with intracellular recordings in perfused explants [38]. Approximately 60% of SON cells respond to low concentrations (30–50 μ M) of GABA with a depression in spontaneous activity, prominent increase in membrane chloride conductance (10–50 nS) and membrane potential hyperpolarization. These GABA-induced potentials demonstrate reversal at approximately -80 mV, similar to that for the reversal of the DBB-evoked IPSP. At higher concentrations the membrane potential change is biphasic with the development of a later depolarization. A GABA-A agonist muscimol produces similar potential and conductance changes but is approximately 20-fold more potent than GABA. The slope of log-log plots of ligand-induced conductances as a function of drug concentration is approximately 2 for both GABA and muscimol, indicating that two molecules combine with the

FIG. 7. Data (from [38]) illustrate intracellular recordings with K-acetate filled micropipettes from two SON MNCs briefly exposed to GABA and muscimol. Hyperpolarizing current pulses were applied to monitor membrane conductance which increases markedly at the peak of the drug-induced responses. Action potential amplitudes are truncated. In A, both GABA and muscimol depress membrane excitability accompanied by hyperpolarization and reduced membrane resistance. In B, from another cell, the suppressant action on firing frequency is accompanied by membrane depolarization in both control media (1) and in the presence of tetrodotoxin (2).

receptor to activate the chloride ionophore. It is of interest that a population of SON neurons requires a higher concentration of GABA ($>100 \mu\text{M}$) to obtain a prominent increase in membrane conductance but accompanied by a membrane depolarization and a GABA reversal potential 10–36 mV more positive than the IPSP reversal potential. This perplexing response has several explanations including chloride influx at dendritic rather than somatic GABA receptors, or the existence of another ionic conductance.

CHEMORECEPTOR INPUT TO MNCs

In contrast to the baroreceptor-induced depression in excitability of hypothalamic MNCs, MNCs are excited by chemoreceptor activation. More specifically, bilateral occlusion of the common carotid arteries, which is likely to activate carotid body chemoreceptors and/or unload carotid sinus baroreceptors, selectively elevates plasma vasopressin and prompts an abrupt and selective onset in firing of phasically-active, i.e., putative vasopressin-secreting MNCs [16]. Subsequent studies have ascertained that this chemoreceptor input is ipsilateral and involves a relay in the anterior hypothalamic-medial preoptic area [17]. The response to carotid sinus chemoreceptor stimulation is consistently abolished by ipsilateral lesions in the ventrolateral medulla, site of a prominent noradrenergic innervation of vasopressin-secreting MNCs [2,17]. It is perhaps more than coincidental that both carotid body chemoreceptor activation and electrical stimulation of the A1 noradrenergic neurons in the ventrolateral medulla selectively excite hypothalamic vasopressin-secreting MNCs. Hence, it is possible that chemoreceptor input to MNCs is relayed directly through the ventrolateral medulla A1 cell group.

OTHER CARDIOVASCULAR INPUTS TO MNCs

An increase in circulating angiotensin II (AII) can produce a rise in arterial blood pressure [3]. Normally, this would tend to activate baroreceptors and subsequently provoke a depression of MNC excitability. However, in the rat, this procedure induces a progressive rise in the excitability of all MNCs [14]. In part, this reflects the ability for

FIG. 8. Schematic to illustrate some of the cardiovascular related inputs to SON and PVN oxytocin and vasopressin-secreting MNCs. On the left, circulating angiotensin II acts on a circumventricular structure, the subfornical organ (SFO), which in turn activates oxytocin and vasopressin MNCs, possibly through a central angiotensinergic circuitry. In the center, peripheral baroreceptor activation provides input to the diagonal band of Broca (DBB) and many other sites through poorly defined pathways that may include nucleus tractus solitarius (NTS), locus coeruleus (LC) and parabrachial nucleus (PBN). It is now proposed that DBB neurons act on a GABAergic interneuron in the perinuclear zone (PNZ) to depress AVP cells selectively. On the right, chemoreceptor activation is likely to send inputs through the NTS to caudal ventrolateral medulla (CVLM) A1 noradrenergic cells. These then may directly activate AVP cells through an ascending noradrenergic input that contains a coexisting neuropeptide NPY.

AII to alter baroreceptor input at the level of the nucleus tractus solitarius and at the same time to activate neurons in a circumventricular structure, the subfornical organ (SFO). SFO neurons project both directly and indirectly to MNCs, producing excitation of both vasopressin- and oxytocin-secreting MNCs [43] and release of corresponding hormones into the plasma [13]. Certain SFO neurons demonstrate angiotensin-like immunoreactivity [32] and preliminary data suggest that angiotensin may be a transmitter in this central circuitry to MNCs [22].

COMMENT

Present electrophysiological studies in rat clearly indicate that stimuli of cardiovascular origin potently exert an influence on the excitability of a variety of hypothalamic neurosecretory neurons. This presentation has emphasized hypothalamic MNCs secreting vasopressin and oxytocin into the plasma (Fig. 8). Although CNS pathways conveying baroreceptor and chemoreceptor influences to MNCs require more precise definition, it would appear that GABA and noradrenaline are excellent transmitter candidates to mediate the respective depression and activation of vasopressin-secreting MNCs from these peripheral sources. In the noradrenergic projection from A1, a coexisting neuropeptide NPY may also have a contributing influence in regulating MNC excitability [10]. Equally intriguing is the potential relationship between circulating angiotensin and central angiotensin in controlling hormone release from the neurohypophysis.

ACKNOWLEDGEMENT

The research was supported by the Quebec Heart Association and Canadian Medical Research Council. We thank Gwen Peard for typing the manuscript.

REFERENCES

1. Arnould, E.; Cirino, M.; Layton, B. S.; Renaud, L. P. Contrasting actions of amino acids, acetylcholine, noradrenaline and leucine enkephalin on the excitability of supraoptic vasopressin-secreting neurons. *Neuroendocrinology* 36:187-196; 1983.
2. Banks, D.; Harris, M. C. Lesions of the locus coeruleus abolish baroreceptor-induced depression of supraoptic neurones in the rat. *J. Physiol.* 355:383-398; 1984.
3. Bickerton, R. K.; Buckley, J. P. Evidence for a central mechanism in angiotensin-induced hypertension. *Proc. Soc. Exp. Biol. Med.* 106:834-836; 1961.
4. Bicknell, R. J.; Leng, G. Relative efficiency of neural firing patterns for vasopressin release in vitro. *Neuroendocrinology* 33:295-299; 1981.
5. Bioulac, B.; Gaffori, O.; Harris, M.; Vincent, J.-D. Effects of acetylcholine, sodium glutamate and GABA on the discharge of supraoptic neurons in the rat. *Brain Res.* 154:159-162; 1978.
6. Bourque, C. W. Intrinsic features and control of phasic burst onset in magnocellular neurosecretory cells. In: Ciriello, J.; Calaresu, F. R.; Renaud, L. P.; Polosa, C., eds. *Organization of the autonomic nervous system: central and peripheral mechanisms*. New York: A. R. Liss; 1987:387-396.
7. Brashear, H. L.; Zaborszky, L.; Heimer, L. Distribution of gabaergic and cholinergic neurons in the rat diagonal band. *Neuroscience* 17:439-451; 1986.
8. Cazalis, M.; Dayanithi, G.; Nordmann, J. J. The role of patterned burst and interburst interval on the excitation-coupling mechanism in the isolated rat neural lobe. *J. Physiol.* 369:45-60; 1985.
9. Day, T. A.; Ferguson, A. V.; Renaud, L. P. Facilitatory influence of noradrenergic afferents on the excitability of rat paraventricular nucleus neurosecretory cells. *J. Physiol.* 355:237-249; 1984.
10. Day, T. A.; Jhamandas, J. H.; Renaud, L. P. Comparison between the actions of avian pancreatic polypeptide, neuropeptide Y and norepinephrine on the excitability of rat supraoptic vasopressin neurons. *Neurosci. Lett.* 62:181-185; 1985.
11. Day, T. A.; Renaud, L. P. Electrophysiological evidence that noradrenergic afferents selectively facilitate the activity of supraoptic vasopressin neurons. *Brain Res.* 303:233-240; 1984.
12. Dreifuss, J. J.; Kalnins, I.; Kelly, J. S.; Ruf, K. B. Action potentials and release of neurohypophysial hormones in vitro. *J. Physiol.* 215:805-817; 1971.
13. Ferguson, A. V. The subfornical organ: a central integration in the control of neurohypophysial hormone secretion. In: Ciriello, J.; Calaresu, F. R.; Renaud, L. P.; Polosa, C., eds. *Organization of the autonomic nervous system: central and peripheral mechanisms*. New York: A. Liss; 1987:435-445.
14. Ferguson, A. V.; Renaud, L. P. Systemic angiotensin acts at subfornical organ to facilitate activity of neurohypophysial neurons. *Am. J. Physiol.* 251:R712-R717; 1986.
15. Gainer, H.; Wolfe, S. A.; Jr., Obaid, A. L.; Salzberg, B. M. Action potentials and frequency-dependent secretion in the mouse neurohypophysis. *Neuroendocrinology* 43:557-563; 1986.
16. Harris, M. C. Effects of chemoreceptor and baroreceptor stimulation on the discharge of hypothalamic supraoptic neurones in rats. *J. Endocrinol.* 82:115-125; 1979.
17. Harris, M. C.; Ferguson, A. V.; Banks, D. The afferent pathway for carotid body chemoreceptor input to the hypothalamic supraoptic nucleus in the rat. *Pflugers Arch.* 400:80-87; 1984.
18. Iijima, K.; Kojima, N. 4-aminobutyrate: 2-oxoglutarate transaminase-containing neurons in the perinuclear zone of the rat supraoptic nucleus. *Acta Histochem.* 79:211-221; 1986.
19. Jhamandas, J. H.; Renaud, L. P. Diagonal band neurons may mediate arterial baroreceptor input to hypothalamic vasopressin-secreting neurons. *Neurosci. Lett.* 65:214-218; 1986.
20. Jhamandas, J. H.; Renaud, L. P. A γ -aminobutyric-acid-mediated baroreceptor input to supraoptic vasopressin neurones in the rat. *J. Physiol.* 381:595-606; 1986.
21. Jhamandas, J. H.; Renaud, L. P. Neurophysiology of a central baroreceptor pathway projecting to hypothalamic vasopressin neurons. *Can. J. Neurol. Sci.* 14:17-24; 1987.
22. Jhamandas, J. H.; Renaud, L. P. Saralasin diminishes subfornical organ-evoked excitation of hypothalamic supraoptic neurosecretory neurons. *Can. J. Physiol. Pharmacol.* 65:Axvii; 1987.
23. Jhamandas, J. H.; Renaud, L. P. Bicuculline blocks an inhibitory baroreflex input to supraoptic vasopressin neurons. *Am. J. Physiol.* 252:R947-R952; 1987.
24. Jhamandas, J. H.; Rogers, J.; Buijs, R. M.; Renaud, L. P. An ultrastructural characterization of a GABA-mediated input to rat supraoptic nucleus (SON) from the diagonal band of Broca (DB). *Soc. Neurosci. Abstr.* 13:1130; 1987.
25. Kannan, H.; Osaka, T.; Kasai, M.; Okuya, S.; Yamashita, H. Electrophysiological properties of neurons in the caudal ventrolateral medulla projecting to the paraventricular nucleus of the hypothalamus in rats. *Brain Res.* 376:342-350; 1986.
26. Kannan, H.; Yagi, K.; Sawaki, Y. Pontine neurones: electrophysiological evidence of mediating carotid baroreceptor inputs to supraoptic neurones in rats. *Exp. Brain Res.* 42:362-370; 1981.
27. Kannan, H.; Yamashita, H. Electrophysiological study of paraventricular nucleus neurons projecting to the dorsomedial medulla and their response to baroreceptor stimulation in rats. *Brain Res.* 279:31-40; 1983.
28. Kannan, H.; Yamashita, H. Connections of neurons in the region of the nucleus tractus solitarius with the hypothalamic paraventricular nucleus: their possible involvement in the neural control of the cardiovascular system in rats. *Brain Res.* 329:205-212; 1985.
29. Kasai, M.; Osaka, T.; Inenaga, K.; Kannan, H.; Yamashita, H. γ -aminobutyric acid antagonist blocks baroreceptor-activated inhibition of neurosecretory cells in the hypothalamic paraventricular nucleus of rats. *Neurosci. Lett.* 81:319-324; 1987.
30. Knuepfer, M. M.; Gebhart, G. F.; Brody, M. J. Effects of baroreceptor activation on single unit activity of the anteroventral third ventricle region of the rat. *Neurosci. Lett.* 56:79-85; 1985.
31. Lincoln, D. W.; Wakerley, J. B. Electrophysiological evidence for the activation of supraoptic neurones during the release of oxytocin. *J. Physiol.* 242:533-554; 1974.
32. Lind, R. W.; Swanson, L. W.; Ganten, D. Angiotensin II immunoreactivity in the neural afferents and efferents of the subfornical organ of the rat. *Brain Res.* 321:209-215; 1984.
33. McAllen, R. M.; Blessing, W. W. Neurons (presumably A₁ cells) projecting from the caudal ventrolateral medulla to the region of the supraoptic nucleus respond to baroreceptor inputs in the rabbit. *Neurosci. Lett.* 73:247-252; 1987.
34. Moore, S. D.; Guyenet, P. G. An electrophysiological study of the forebrain projection of nucleus commissuralis: preliminary identification of presumed A2 catecholaminergic neurons. *Brain Res.* 263:211-222; 1983.
35. Poulain, D. A.; Wakerley, J. B. Electrophysiology of hypothalamic magnocellular neurones secreting oxytocin and vasopressin. *Neuroscience* 7:773-808; 1982.
36. Randle, J. C. R.; Bourque, C. W.; Renaud, L. P. Characterization of spontaneous and evoked inhibitory postsynaptic potentials in rat supraoptic neurosecretory neurons in vitro. *J. Neurophysiol.* 56:1703-1717; 1986.
37. Randle, J. C. R.; Bourque, C. W.; Renaud, L. P. α_1 -Adrenergic receptor activation depolarizes rat supraoptic neurosecretory neurons in vitro. *Am. J. Physiol.* 251:R569-R574; 1986.
38. Randle, J. C. R.; Renaud, L. P. Actions of γ -aminobutyric acid on rat supraoptic nucleus neurosecretory neurones in vitro. *J. Physiol.* 387:629-647; 1987.
39. Renaud, L. P. Magnocellular neuroendocrine neurons: update on intrinsic properties, synaptic inputs and neuropharmacology. *Trends Neurosci.* 10:498-502; 1987.

40. Renaud, L. P. Electrophysiology of a peptidergic neuron—the hypothalamic magnocellular neurosecretory cell. In: Avoli, M.; Reader, T.; Dykes, R. W.; Gloor, P., eds. *Neurotransmitters and cortical function*. New York: Plenum Press; 1988, in press.
41. Renaud, L. P.; Bourque, C. W.; Day, T. A.; Ferguson, A. V.; Randle, J. C. R. Electrophysiology of mammalian hypothalamic supraoptic and paraventricular neurosecretory cells. In: Poisner, A.; Trifaro, J., eds. *The electrophysiology of the secretory cell*. Amsterdam: Elsevier; 1985:165–194.
42. Renaud, L. P.; Tang, M.; McCann, J. J.; Stricker, E. M.; Verbalis, J. G. Cholecystokinin and gastric distention activate oxytocinergic cells in rat hypothalamus. *Am. J. Physiol.* 253:R661–R665; 1987.
43. Sgro, S.; Ferguson, A. V.; Renaud, L. P. Subfornical organ-supraoptic nucleus connections: an electrophysiological study in the rat. *Brain Res.* 303:7–13; 1984.
44. Silverman, A. J.; Hoffman, D. L.; Zimmerman, E. A. The descending afferent connections of the paraventricular nucleus of the hypothalamus (PVN). *Brain Res. Bull.* 6:47–61; 1981.
45. Swaab, D. F.; Pool, C. W.; Nijveldt, F. Immunofluorescence of vasopressin and oxytocin in the rat hypothalamo-neurohypophyseal system. *J. Neural Transm.* 36:195–215; 1975.
46. Swanson, L. W.; Cowan, W. M. The connections of the septal region in the rat. *J. Comp. Neurol.* 186:621–655; 1979.
47. Theodosis, D. T.; Paut, L.; Tappaz, M. L. Immunocytochemical analysis of the GABAergic innervation of oxytocin- and vasopressin-secreting neurons in the rat supraoptic nucleus. *Neuroscience* 19:207–222; 1986.
48. Thornton, S. N.; de Beaurepaire, R.; Nicolaidis, S. Electrophysiological investigation of cells in the region of the anterior hypothalamus firing in relation to blood pressure and volaemic changes. *Brain Res.* 299:1–7; 1984.
49. Tribollet, E.; Armstrong, W. E.; Dubois-Dauphin, M.; Dreifuss, J. J. Extrahypothalamic afferent inputs to the supraoptic nucleus area of the rat as determined by retrograde and anterograde tracing techniques. *Neuroscience* 15:135–138; 1985.
50. Van Den Pol, A. Dual ultrastructural localization of two neurotransmitter-related antigens: colloidal gold-labeled neurophysin-immunoreactive supraoptic neurons receive peroxidase-labeled glutamate decarboxylase- or gold-labeled GABA-immunoreactive synapses. *J. Neurosci.* 5:2940–2954; 1985.
51. Vandesande, F.; Dierickx, K. Identification of the vasopressin producing and of the oxytocin producing neurons in the hypothalamic neurosecretory system of the rat. *Cell Tissue Res.* 164:153–162; 1975.
52. Yamashita, H.; Inenaga, K.; Kannan, H. Depolarizing effect of noradrenaline on neurons of the rat supraoptic nucleus in vitro. *Brain Res.* 405:348–352; 1987.
53. Yamashita, H.; Kannan, H.; Inenaga, K.; Koizumi, K. Role of neurons in the supraoptic and paraventricular nuclei in cardiovascular control. In: Cross, B. A.; Leng, G., eds. *The neurohypophysis: Structure, function and control*. Amsterdam: Elsevier; 1983:459–468.