

International Conference on the Neurohypophysis, Key Biscayne, Florida  
November 14–19, 1976

---

# Neurohypophysis

Editors

*A.M. Moses*, Syracuse, N.Y., and *L. Share*, Memphis, Tenn.

62 figures and 20 tables, 1977



---

S. Karger · Basel · München · Paris · London · New York · Sydney

---

*Other publications in this field*

**Brain-Endocrine Interaction II. The Ventricular System in Neuroendocrine Mechanisms**

2nd International Symposium on Brain-Endocrine Interaction, Shizuoka 1974

Editors: *K.M. Knigge* and *D.E. Scott*, Rochester, N.Y.; *H. Kobayashi*, Miura-shi, and *S. Ishii*, Tokyo

X + 406 p., 181 fig., 25 tab., 2 cpl., 1975. ISBN 3-8055-2176-6

**Brain-Endocrine Interaction III. Neural Hormones and Reproduction**

3rd International Symposium on Brain-Endocrine Interaction, Würzburg 1977

Editors: *D.E. Scott*, Rochester, N.Y.; *G.P. Kozlowski*, Fort Collins, Colo., and *A. Weindl*, München

approx. 380 p., 179 fig., 15 tab., 1 cpl., 1978. ISBN 3-8055-2798-5

Bayarische  
Staatsbibliothek  
München

---

**Cataloging in Publication**

International Conference on the Neurohypophysis, 2d, Key Biscayne, Fla., 1976

Neurohypophysis: International Conference on the Neurohypophysis, Key Biscayne, Fla., November 14-19, 1976

Editors, A.M. Moses, L. Share. - Basel; New York: Karger, 1977.

1st conference called Symposium on the Neurohypophysis.

I. Pituitary Gland, Posterior - congresses I. Moses, A.M., ed. II. Share, L., ed.

III. Symposium on the Neurohypophysis, University of Bristol, 1956 IV. Title

W3 IN19483N 1976n/ WK 520 I58 1976n

ISBN 3-8055-2664-4

---

All rights reserved.

No part of this publication may be translated into other languages, reproduced or utilized in any form or by any means, electronic or mechanical, including photocopying, recording, microcopying, or by any information storage and retrieval system, without permission in writing from the publisher.

- © Copyright 1977 by S. Karger AG, 4011 Basel (Switzerland), Arnold-Böcklin-Strasse 25  
Printed in Switzerland by Thür AG Offsetdruck, Pratteln  
ISBN 3-8055-2664-4

---

# Contents

Organizing Committee	VII
Participants	VII
Preface	IX
<i>Sawyer, W.H. and Pang, P.K.T.</i> (New York, N.Y.): Evolution of Neurohypophysial Hormones and Their Functions	1
<i>Manning, M. and Sawyer, W.H.</i> (New York, N.Y.): Structure-Activity Studies on Oxytocin and Vasopressin 1954-1976. From Empiricism to Design	9
<i>Zimmerman, E.A. and Defendini, R.</i> (New York, N.Y.): Hypothalamic Pathways Containing Oxytocin, Vasopressin and Associated Neurophysins	22
<i>Pickering, B.T. and McPherson, M.A.</i> (Bristol): Progress in the Study of Biosynthesis and Transport in the Neurohypophysial System	30
<i>North, W.G.; Morris, J.F.; La Rochelle, F.T., and Valtin, H.</i> (Hanover, N.H.): Enzymatic Interconversions of Neurophysins. The Nature of Enzyme(s) within Neurosecretory Granules of the Neurohypophysis	43
<i>Schlesinger, D.H.; Walter, R., and Audhya, T.K.</i> (Chicago, Ill.): Separation of Rat Neurophysins I and II by Ion-Exchange Chromatography and Their Partial Amino Acid Sequences	53
<i>Morris, J.F.; Sokol, H.W., and Valtin, H.</i> (Hanover, N.H.): One Neuron - One Hormone? Recent Evidence from Brattleboro Rats	58
<i>Hayward, J.N.</i> (Chapel Hill, N.C.): Hypothalamic Magnocellular Neuroendocrine Cell Activity and Neurohypophysial Hormone Release	67
<i>Dyball, R.E.J.; Wakerley, J.B.; Poulain, D.A., and Brimble, M.J.</i> (Babraham, Cambs.): Electrophysiological Determinants of Neurosecretion	78
<i>Theodosis, D.T. and Dreifuss, J.J.</i> (Geneva): Ultrastructural Evidence for Exo-Endocytosis in the Neurohypophysis	88
<i>Thorn, N.A.; Russell, J.T.; Dahl, G., and Gratzl, M.</i> (Copenhagen): Studies of the Mechanism of Antidiuretic Hormone Release	95
<i>Lincoln, D.W.; Clarke, G.; Mason, C.A., and Dreifuss, J.J.</i> (Bristol): Physiological Mechanisms Determining the Release of Oxytocin in Milk Ejection and Labour	101
<i>Mathison, R. and Lederis, K.</i> (Calgary): Cyclic Nucleotides in the Hypothalamo-Neurohypophysial System and the Release of Vasopressin	110
<i>Andersson, B. and Olsson, K.</i> (Stockholm): Evidence for Periventricular Sodium-Sensitive Receptors of Importance in the Regulation of ADH Secretion	118

---

# International Conference on the Neurohypophysis

Key Biscayne, Fla., November 14–19, 1976

## *Organizing Committee*

Barry Cross, Cambridge  
Myron Miller, Syracuse, N.Y.  
Arnold M. Moses, Syracuse, N.Y.

Wilbur H. Sawyer, New York, N.Y.  
Berta Scharrer, Bronx, N.Y.  
Leonard Share, Memphis, Tenn.

## *Participants*

Charles F. Abbound, Rochester, Minn.  
Bengt Andersson, Stockholm  
David M. Brennan, Indianapolis, Ind.  
Neil Breslau, Syracuse, N.Y.  
Maurice Bugariu, East Northport, N.Y.  
Mei-Fang Cheng, Newark, N.J.  
John R. Claybaugh, San Francisco, Calif.  
Richard Coulson, Syracuse, N.Y.  
Joan T. Crofton, Memphis, Tenn.  
Barry Cross, Cambridge  
Richard Defendini, New York, N.Y.  
H. Dieter Dellmann, Ames, Iowa  
David de Wied, Utrecht  
Thomas P. Dousa, Rochester, Minn.  
J.J. Dreifuss, Geneva  
Richard E.J. Dyball, Cambridge  
Craig Ferris, Ossining, N.Y.  
George R. Flouret, Deerfield, Ill.  
Mary Louise Forsling, London  
Christine M. Gregg, Rochester, N.Y.  
Jaya Haldar, New York, N.Y.  
James N. Hayward, Chapel Hill, N.C.  
Alan B. Huellmantel, Pittsburgh, Pa.  
Stefanie M. Huk, South Orange, N.J.

Serge Jard, Paris  
David P. Jennings, Stillwater, Okla.  
Lanny Keil, Moffett Field, Calif.  
Frederick T. La Rochelle, Hanover, N.H.  
Karl Lederis, Calgary  
Dennis W. Lincoln, Bristol  
Richard L. Malvin, Ann Arbor, Mich.  
Maurice Manning, Toledo, Ohio  
Ronald Mathison, Calgary  
Myron Miller, Syracuse, N.Y.  
John F. Morris, Bristol  
Arnold M. Moses, Syracuse, N.Y.  
William G. North, Hanover, N.H.  
Peter K.T. Pang, Lubbock, Tex.  
David Pearson, Davis, Calif.  
Brian T. Pickering, Bristol  
Vladimir Pliska, Zurich  
Ralph Rabkin, Memphis, Tenn.  
Raymond V. Randall, Rochester, Minn.  
Al Reaves, Chapel Hill, N.C.  
John S. Roberts, Natick, Mass.  
Alan G. Robinson, Pittsburgh, Pa.  
Leonard J. Robinson, Lowell, Mass.  
Alan B. Rothballer, New York, N.Y.

- and contraluminal plasma membranes as a tool for the analysis of transport processes and hormone action; in *Bolis, Hoffman and Leaf* Membranes and disease, pp. 331–343 (Raven Press, Hewlett 1976).
- 21 *Evers, C.; Murer, H., and Kinne, R.*: Unpublished data; cited in *Kinne and Schwartz* The asymmetrical distribution of renal epithelial cell membrane function in the action of antidiuretic hormone and parathyroid hormone; in *Andreoli, Grantham and Rector* Fluid osmolality (Am. Physiol. Soc., in press).
  - 22 *Kinne, R.; Shlatz, L.J.; Kinne-Saffran, E., and Schwartz, I.L.*: Distribution of membrane-bound cyclic AMP-dependent protein kinase in plasma membranes of cells of the kidney cortex. *J. Membr. Biol.* 24: 145–159 (1975).
  - 23 *Kirchberger, M.A.; Schwartz, I.L., and Walter, R.*: Cyclic 3',5'-AMP dependent protein kinase activity in toad bladder epithelium. *Proc. Soc. exp. Biol. Med.* 140: 657–660 (1972).
  - 24 *Kyte, J.*: Immunoferritin determination of the distribution of (Na<sup>+</sup>+K<sup>+</sup>)ATPase over the plasma membranes of renal convoluted tubules. II. Proximal segment. *J. Cell Biol.* 68: 304–318 (1976).
  - 25 *Ripoche, P.A.; Huang, C.J., and Schwartz, I.L.*: cAMP-dependent protein kinases in the canine kidney inner medulla. *Fed. Proc. Fed. Am. Socs exp. Biol.* (in press).
  - 26 *Schmidt, U. and Dubach, U.C.*: Na K stimulated adenosinetriphosphatase: intracellular localization within the proximal tubule of the rat nephron. *Pflügers Arch.* 330: 265–270 (1971).
  - 27 *Schwartz, I.L.*: Structure-activity relationships as a tool for the analysis of sequential events in the action of neurohypophyseal hormones on membrane permeability; in *Margoulies and Greenwood* Structure-activity relationships of protein and polypeptide hormones. *Int. Congr. Ser. No. 241, part I*, pp. 31–37 (Excerpta Medica, Amsterdam 1971).
  - 28 *Schwartz, I.L. and Walter, R.*: Factors influencing the reactivity of the toad bladder to the hydro-osmotic action of vasopressin. *Am. J. Med.* 42: 769–776 (1967).
  - 29 *Schwartz, I.L.; Shlatz, L.J.; Kinne-Saffran, E., and Kinne, R.*: Target cell polarity and membrane phosphorylation in relation to the mechanism of action of antidiuretic hormone. *Proc. natn. Acad. Sci. USA* 71: 2595–2599 (1974).
  - 30 *Shlatz, L.J.; Schwartz, I.L.; Kinne-Saffran, E., and Kinne, R.*: Distribution of parathyroid hormone-stimulated adenylate cyclase in plasma membranes of cells of the kidney cortex. *J. Membr. Biol.* 24: 131–144 (1975).
  - 31 *Szoka, F.C., jr. and Ettinger, M.J.*: Electrophoretic analyses of phosphorylated kidney membranes in the presence of 3',5'-cyclic adenosine monophosphate and antidiuretic hormone. *Fed. Proc. Fed. Am. Socs exp. Biol.* 34: 543 (1975).
  - 32 *Walton, K.G.; DeLorenzo, R.J.; Curran, P.F., and Greengard, P.*: Regulation of protein phosphorylation and sodium transport in toad bladder. *J. gen. Physiol.* 65: 153–177 (1975).

## Studies of the Mechanism of Antidiuretic Hormone Release

*N.A. Thorn, J.T. Russell, G. Dahl and M. Gratzl*

Institute of Medical Physiology C, University of Copenhagen, Copenhagen, and  
Fachbereich Theoretische Medizin, Universität des Saarlandes, Homburg

### *Introduction*

In recent years, a considerable amount of information has been collected concerning the role of calcium in the release of neurohypophysial hormones. Also, the possible role of ATP has been studied to some extent. The hypothalamo-neurohypophysial system in these respects can probably be taken as a model system for neurosecretion.

The authors here take the liberty of mainly reporting new results of their own. Several reviews of previous experiments on these problems have been published during the past few years (1, 8, 9, 18–21).

### *Movement of Calcium into the Neurosecretory Nerve Endings on Stimulation and Reestablishment of a Normal Low Intracellular Concentration of Free Calcium*

It seems now well established that on stimulation of the neurosecretory cells a trigger amount of calcium moves over the cell membrane, probably through special calcium 'channels' (7, 10–12, 16, 17) and induces release of hormone, probably by exocytosis. The intracellular concentration of free calcium is presumably very low (of the order of  $10^{-7} M$ ) (18). For this system to be able to work, there must exist mechanisms which normalize the increased intracellular concentration of free calcium after stimulation. These processes involve transport of calcium out over the cell membrane, probably at least some of it being dependent on sodium transport the opposite way. The problem of the existence of a calcium 'pump' of the type known, e.g. in erythrocyte cell membrane, has not yet been solved. In addition, various sequestering mechanisms are able temporarily to reduce the axoplasmic concentration of free calcium. This especially involves mitochondria, which have an ATP-dependent calcium uptake (13).

We have previously (21) found that isolated secretory granules contain a high concentration of calcium. The granules were prepared at low temperature and the calcium concentration may be higher if the preparation is made at more normal temperatures. We have demonstrated an ATP-dependent uptake of calcium by such granules. It was small when expressed per unit granule protein (13). If one, however, expresses it per unit granule membrane protein, it naturally becomes much higher. The granule preparation only took up calcium for a short time, then the granules burst. This is probably due to an ATPase-dependent proton (and chloride) transport into the granules with ensuing osmotic effect. Such a mechanism has been suggested for catecholamine-containing granules (2).

Sequestration might also involve binding to endoplasmic reticulum, e.g. a binding to proteins of the same character as those which have been demonstrated to be present in the sarcoplasmic reticulum.

Recently, a number of analogies have been pointed out between stimulus-secretion coupling and stimulus-contraction coupling. Muscle biochemistry is much more advanced in these respects than secretion biochemistry (1, 5). It would seem appropriate to apply some of the findings and ideas of muscle biochemistry to secretion biochemistry since so many functional analogies can be drawn. This has recently been done concerning calcium-binding proteins. Striated muscle contains proteins with a high affinity for calcium (5) which seem to have a clear function in contraction. In addition, there are also present in muscle tissue proteins with a lesser affinity, such as modulators of cyclic nucleotide phosphodiesterase and inhibitors of such modulators (6, 23).

We have recently started a series of studies on isolation of calcium-binding proteins from ox neurohypophyses. So far, we have isolated one in a high degree of purity from the soluble fraction (14). Its molecular weight was approximately 15,000. The purification procedure involved ammonium sulphate fractionation, DEAE-cellulose chromatography and gel filtration on Sephadex G-100 and Sephadex G-50. Conventional and SDS polyacrylamide gel electrophoresis demonstrated it to be a protein distinct from the S-100 protein and the soluble hormone-binding proteins (neurophysins) abundant in the neurohypophysis. It appears to be the only calcium-binding protein in the soluble part of the homogenate. It has an apparent  $K_{\text{diss}}$  for calcium of  $1.1 \times 10^{-5} M$  (at 22 °C) and a binding capacity of 2 mol of calcium per mole of protein. This protein may be identical to the calcium-dependent modulator of cyclic nucleotide phosphodiesterase, isolated by Lin *et al.* (6) from bovine brain. It may also be identical to similar proteins isolated from adrenal medulla and the electroplax of the electric eel. From the microsomal fraction from ox neurohypophyses, we have (after solubilization) isolated a calcium-binding protein which in some respects had somewhat similar properties (binding capacity and dissociation constant) as the one isolated from the soluble fraction. These proteins may be identical. Mem-

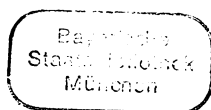
brane-bound calcium-stimulated ATPases have been isolated from ox neurohypophyses (22). It would appear unlikely that the 2 proteins with molecular weights of approximately 15,000 are fragments of such ATPases. However, from the deoxycholate fraction also a protein with a molecular weight of approximately 68,000 was isolated. Calcium-binding proteins were also present in the deoxycholate *insoluble* fraction. The latter group of calcium-binding proteins all need further characterization. There may be proteins with a really high affinity for calcium among them. These proteins might serve crucial functions in the release process, e.g. stimulated by the trigger amounts of calcium entering the cells.

### *Mechanism of Action of Trigger Calcium*

Recently, Drs. *Dahl and Gratzl* from Homburg, West Germany, and Dr. *Russell* and I, in our laboratory, have studied problems of cation specificity and concentration dependence of calcium-induced fusion of isolated neurosecretory vesicles from ox neurohypophyses. The essential features of the studies (27) were that isolated secretory vesicles were incubated in a very simple medium (Na-cacodylate 10 mM, pH 7.0; sucrose 250 mM, and EGTA 1 mM) and that the effects of increasing the calcium concentration in the medium from less than  $10^{-7}$  to  $10^{-3}$  M on fusion of granules were studied. Fusion was studied by freeze fracture electron microscopy which especially demonstrates aspects of the membranes in the preparations. With increasing calcium concentration, an increase of fusion was observed with a half-maximal effect between  $10^{-6}$  and  $10^{-5}$  M calcium. The changes could not be produced by magnesium or strontium. These morphological changes of neurosecretory vesicles were thus similar to changes demonstrated by *Dahl and Gratzl* (3, 25, 26) in other secretory tissues, such as pancreatic islets, liver and adrenal medulla. In the present experiments in addition, it was shown that in parallel to the increased fusion there was an increased release of vasopressin to the medium, with a similar concentration dependence and ion specificity. The vesicle preparations contained sheets of plasma membrane and the increased hormone release observed might occur by exocytosis-like processes.

### *Studies of ATP-Induced Emptying of Neurosecretory Vesicle Content of Vasopressin*

Another important aspect of vasopressin release is the fact that it has been reported to be dependent on energy. This was already found by *Poisner and Douglas* (4) in 1968 in experiments where isolated neurosecretory granules released vasopressin, oxytocin, and protein on exposure to ATP. They also ob-





served ATP-splitting activity associated with a purified granule preparation. They suggested that ATP and ATPase may participate in the processes leading to the release of hormones from the neurohypophysis. *Warberg and Thorn* (24) repeated the experiments of *Douglas and Poisner* and found essentially the same results. However, they stressed the fact that the apparent dependence of release on energy might involve other processes than the release step itself, i.e. that energy was necessary to keep the activation machinery intact.

*Vilhardt and Hope* (22), on the other hand, have been unable to find ATPase activity associated with the neurosecretory granules. *Russell and Thorn* (15) have recently investigated the effect of ATP on isolated secretory granules with a view to try to clarify whether the ATP-induced release of granule contents is due to a membrane instability caused by ATP or an enzyme-mediated effect.

It was demonstrated that isolated neurohypophysial secretory vesicles released their content of vasopressin in the presence of ATP. The release was nearly complete. It occurred with a half-maximal ATP concentration of 0.25 mM. Calcium was not necessary for the effect. This fact alone shows that this specific type of release is of a different kind than normal release which requires calcium. ADP, AMP, and ITP were shown to mimic the effect of ATP. This fact suggests that ATPase is not involved in this release and that it is rather due to an instability of the vesicle membrane caused by the nucleotides. On the other hand, some involvement of ATPase is possible. The background for this suggestion was the following: The ATP effect was dependent on magnesium. AMP-PCP, a non-metabolizable ATP analog, inhibited the ATP-induced granule emptying. Although this effect might be explained via binding of magnesium by AMP-PCP, this was not sufficient to explain the inhibition. These findings therefore might go along with the hypothesis that there is some ATP splitting in these experiments. This suggestion was actually supported by experiments on phosphorylation of the granule membranes. Utilizing the property of ATP to selectively render the secretory granules lighter, a method was developed to obtain granule 'ghosts' in a highly purified form. These purified membrane ghosts were shown to be phosphorylated in the presence of ATP. After the conclusion of these experiments, an article on ATP-evoked catecholamine release from catecholaminergic granules was published by *Casey et al.* (2). They suggested that the release is driven by an inwardly directed proton-translocating adenosine triphosphatase. A resulting proton-anion influx causes osmotic lysis of the chromaffin granules. They considered it unlikely that there was any involvement of this process in the normal release process. As previously stated, such a mechanism might well be responsible for part of the release found in our experiments on isolated neurohypophysial secretory granules. It would seem that it must be concluded that so far no clear evidence has been presented that ATP is critically involved in the release process itself.

## References

- 1 *Carafoli, E.; Clementi, F.; Drabikowski, W., and Margreth, A.* (eds): Calcium transport in contraction and secretion (North-Holland, Amsterdam 1975).
- 2 *Casey, R.P.; Njus, D.; Radda, G.R., and Sehr, P.A.*: Adenosine triphosphate-evoked catecholamine release in chromaffin granules. *Biochem. J.* 158: 583–588 (1976).
- 3 *Dahl, G. and Gratzl, M.*: Calcium-induced fusion of isolated secretory vesicles from the islet of Langerhans. *Cytobiology* 12: 344–355 (1976).
- 4 *Poisner, A.M. and Douglas, W.W.*: A possible mechanism of release of posterior pituitary hormones involving ATP and an ATPase in the neurosecretory granules. *Mol. Pharmacol.* 4: 531–540 (1968).
- 5 *Drabikowski, W.; Strzelecka-Golaszewska, H., and Carafoli, E.* (eds): Calcium binding proteins. *Proc. Int. Symp., Jablonna 1973* (Elsevier, Amsterdam 1974).
- 6 *Lin, Y.M.; Liu, Y.P., and Cheung, W.Y.*: Cyclic 3':5'-nucleotide phosphodiesterase. Purification, characterization, and active form of the protein activator from bovine brain. *J. biol. Chem.* 249: 4943–4954 (1974).
- 7 *Müller, J.R.; Thorn, N.A., and Torp-Pedersen, C.*: Effects of calcium and sodium on vasopressin release *in vitro* induced by a prolonged potassium stimulation. *Acta endocr. Copenh.* 79: 51–59 (1975).
- 8 *Poisner, A.M.*: Stimulus-secretion coupling in the adrenal medulla and posterior pituitary gland; in *Ganong and Martini Frontiers in neuroendocrinology*, pp. 33–59 (Oxford University Press, London 1973).
- 9 *Poisner, A.M.*: The role of calcium in neuroendocrine secretion; in *Naftolin, Ryan and Davies Subcellular mechanisms in reproductive neuroendocrinology*, pp. 45–62 (Elsevier, Amsterdam 1976).
- 10 *Robinson, I.C.A.F.; Russell, J.T., and Thorn, N.A.*: Calcium and stimulus secretion coupling in the neurohypophysis. V. The effects of the  $\text{Ca}^{2+}$  ionophores A23187 and X536A on vasopressin release and  $^{45}\text{Ca}^{2+}$  efflux; interactions with sodium and a verapamil analogue (D600). *Acta endocr. Copenh.* 83: 36–49 (1976).
- 11 *Russell, J.T. and Thorn, N.A.*: Calcium and stimulus secretion coupling in the neurohypophysis. I. 45-Calcium transport and vasopressin release in slices from ox neurohypophyses stimulated electrically or by a high potassium concentration. *Acta endocr., Copenh.* 76: 449–470 (1974).
- 12 *Russell, J.T. and Thorn, N.A.*: Calcium and stimulus secretion coupling in the neurohypophysis. II. Effects of lanthanum, a verapamil analogue (D600) and prenylamine on 45-calcium transport and vasopressin release in isolated rat neurohypophyses. *Acta endocr. Copenh.*, 76: 471–487 (1974).
- 13 *Russell, J.T. and Thorn, N.A.*: Adenosine triphosphate-dependent calcium uptake by subcellular fractions from bovine neurohypophyses. *Acta physiol. scand.* 93: 364–377 (1975).
- 14 *Russell, J.T. and Thorn, N.A.*: Isolation and purification of calcium-binding proteins from bovine neurohypophyses. *Biochim. biophys. Acta* 491: 398–408 (1977).
- 15 *Russell, J.T. and Thorn, N.A.*: Adenosine triphosphate-induced emptying of isolated bovine neurohypophysial secretory granule contents (in preparation).
- 16 *Russell, J.T.; Hansen, E.L., and Thorn, N.A.*: Calcium and stimulus-secretion coupling in the neurohypophysis. III.  $\text{Ca}^{2+}$  ionophore (A-23187)-induced release of vasopressin from isolated rat neurohypophyses. *Acta endocr. Copenh.*, 77: 443–450 (1974).
- 17 *Russell, J.T.; Warberg, J., and Thorn, N.A.*: Calcium and stimulus-secretion coupling in the neurohypophysis. IV. Effects of *N*-ethylmaleimide on  $^{45}\text{Ca}^{2+}$  transport and vasopressin release by isolated rat neurohypophyses. *Acta endocr., Copenh.* 77: 691–698 (1974).

- 18 Thorn, N.A.: Role of calcium in secretory processes; in *Thorn and Petersen Secretory mechanisms of exocrine glands*. Alfred Benzon Symp., vol. 8, pp. 305–326 (Munksgaard, Copenhagen 1974).
- 19 Thorn, N.A.; Russell, J.T., and Robinson, I.C.A.F.: Factors affecting intracellular concentration of free calcium ions in neurosecretory nerve endings; in *Carafoli, Clementi and Margreth Calcium transport in contraction and secretion*, pp. 261–269 (North-Holland, Amsterdam 1975).
- 20 Thorn, N.A.; Russell, J.T., and Sunde, D.: Mechanism of cellular antidiuretic hormone release. Proc. 5th Int. Congr. Endocrinol. (in press, 1976).
- 21 Thorn, N.A.; Russell, J.T., and Vilhardt, H.: Hexosamine, calcium and neurophysin in secretory granules and the role of calcium in hormone release. Ann. N.Y. Acad. Sci. 248: 202–217 (1975).
- 22 Vilhardt, H. and Hope, D.B.: Adenosine triphosphatase activity in the neural lobe of the bovine pituitary gland. Biochem. J. 143: 181–190 (1974).
- 23 Wang, J.H. and Desai, R.: A brain protein and its effect on the  $Ca^{2+}$  – and protein modulator-activated cyclic nucleotide phosphodiesterase. Biochem. biophys. Res. Commun. 72: 926–932 (1976).
- 24 Warberg, J. and Thorn, N.A.: *In vitro* studies of the release mechanism for vasopressin in rats. III. Effect of metabolic inhibitors on the release. Acta endocr. Copenh. 61: 415–429 (1969).
- 25 Gratzl, M. and Dahl, G.:  $Ca^{2+}$ -induced fusion of Golgi-derived secretory vesicles isolated from rat liver. FEBS Lett. 62: 142–145 (1976).
- 26 Dahl, G.; Gratzl, M., and Ekerdt, R.: *In vitro* fusion of secretory vesicles isolated from pancreatic B-cells and from the adrenal medulla. J. Cell Biol. 70: 180a (1976).
- 27 Gratzl, M.; Dahl, G.; Russell, J.T., and Thorn, N.A.: Fusion of neurohypophyseal membranes *in vitro*. Biochim. biophys. Acta (in press).

---

## Subject Index

- ACTH 35, 181, 201
- Action potentials
  - paraventricular nuclei 78
  - supraoptic nuclei 78
- Adenylate cyclase
  - ADH 211, 214–217, 220, 227, 228, 237–239, 244
  - catecholamines 216
  - fluoride 217
  - guanylyl 5'-imidodiphosphate 217
  - isoproterenol 146
  - norepinephrine 146
  - PTH 236–238
  - regulation 216
- ADH, *see* Vasopressin
- Adrenal insufficiency
  - neurophysin 139, 140
  - vasopressin 139
- $\beta$ -Adrenergic
  - antidiuretic 145
  - depression 74
- Adrenocortical hormones
  - water excretion 149
- Alkaline phosphatase 238
- Amnesia
  - desglycinamide<sup>9</sup>-lysine<sup>8</sup> vasopressin 201
  - puromycin 203
- Anesthesia
  - antidromic facilitation 74
  - AVP release 128, 129
  - axonal conduction 73
  - diuresis 129
  - oxytocin release 73
  - recurrent collaterals 74
- Angiotensin
  - sodium interaction 121, 123
  - vasopressin release 110
- ATP
  - calcium uptake
    - secretory granules 96
  - magnesium dependence 98
  - neurohypophysis
    - hormone release 95, 98
  - neurosecretory granules 97
- ATPase
  - calcium stimulated 97
  - neurohypophysis
    - hormone release 98
  - proton transport 96, 98
- AVP, *see* Vasopressin
- D*-AVP 12
- AVT
  - effects of
    - antidiuretic 4
    - cardiovascular 5, 6
    - diuretic 4, 5
    - facilitation, self-induced 244
    - glomerular circulation 4, 5
    - intrinsic inhibition, self-induced 244
    - renal 4–6
    - smooth muscle 6
    - vasopressor 3
    - water excretion 4
    - water permeability 4

- AVT (continued)  
  4-glutamine 3  
  neurohypophysial peptide theory 3  
  point mutations 3  
Axon plasmalemma  
  depolarization 88  
Axonal conduction  
  anesthesia, effects of 73
- Baroreceptor pathways 145  
Baroreceptors  
  arterial 131  
  atrial 146  
  carotid sinus 132  
  function 131
- Behavior  
  ACTH 201  
  AVP release 71, 72  
  bradycardia 202  
  cerebrospinal fluid 203  
  desglycinamide<sup>9</sup>-lysine<sup>8</sup> vasopressin  
    201–206  
  *L*-dopa 203  
  LVP 201, 202  
  MSH 201  
  neuropeptides 206  
  nicotine 74  
  oxytocin 205  
  vasopressin 201, 202, 204
- $\beta$ -Blocking drug  
  propranolol 145
- Blood gas tension 128  
Blood pressure 128, 131  
8Br-cyclic AMP 111  
Butorphanol 162
- Caffeine 110  
Calcitonin 238  
Calcium  
  binding protein 96  
  hormone release 85, 95, 110  
  neurosecretory vesicles 97  
  renal microtubular 229
- Carbamazepine 160  
Carboxamidopeptidase 173  
Carotid occlusion  
  effects of 77, 131–133  
Catecholamines  
  vasopressin interaction 146  
Catecholamine-containing granules 96
- Cell polarity  
  hormone action 241
- Cerebral sensory mechanism  
  ADH secretion 120  
  water intake 120
- Cerebrospinal fluid  
  sodium elevation 119
- Chemoreceptors  
  AVP secretion 131, 132, 134
- Chlorpropamide 156, 160  
8-*p*-Chlorophenylthio-cyclic AMP 225
- Cholinergic synapse 74  
Chymotrypsin 169, 172
- Circadian rhythm  
  AVP 71, 72
- Clofibrate 156, 160
- Collateral inhibitory synaptic  
  transmitter 68
- Collecting duct cells 239–241, 243
- Contraluminal membrane  
  ADH, response to 238
- Cyclic AMP  
  catecholamines 146  
  demethylchlortetracycline 164  
  dephosphorylation 241  
  lithium 164  
  oxytocin secretion 110  
  phosphorylation 238, 240, 243  
  renal medulla 141, 146, 220, 221, 240,  
    243  
  vasopressin secretion 110, 115
- Cyclic AMP-cyclic GMP system 114
- Cyclic GMP  
  vasopressin release 110
- Cyclic nucleotides  
  vasopressin release 110
- Cyclo-leu-gly  
  behavioral effect 206, 207
- Cystine aminopeptidase 169, 174
- Cytoplasmic hypertrophy 62  
Cytoplasmic pool 88
- Dale's principle 58  
dDAVP 14, 15, 160, 181, 215, 216  
Deamino-oxytocin 11–13, 16, 17  
Deamino-4-threonine-oxytocin 12  
Demethylchlortetracycline 164  
Dephosphorylation 236  
Deserpidine 181  
Desglycinamide-arginine vasopressin 202

- Desglycinamide-lysine vasopressin 201, 202, 204–206
- Diabetes insipidus
- $\alpha$ -adrenergic stimulation 146
  - $\beta$ -adrenergic stimulation 146
  - causes 153, 159
  - memory 204–206
  - nephrogenic 164
  - rhythmic slow activity 205
- Dibutyl cyclic AMP 111, 238
- dLVP 180
- L*-Dopa 203
- dPTOT 16
- dPVDVP 17
- dTDVP 15
- dVDVP 14, 15, 17
- Electrical membrane properties 68
- Electrical stimulation
- pituitary stalk 78
- Electron-dense granules 64
- Endocytosis 90
- Endopeptidase 177
- Epinephrine 238
- Estrogen-stimulated neurophysin 23, 26, 27, 136, 139
- Exocytosis 88–90, 92
- Ferguson style reflex 107, 108
- Glucocorticoid 149
- Glycerol 120
- 7-Glycine-oxytocin 16
- Gly-NH<sub>2</sub>-releasing enzyme 172
- Henle's loop 211
- Histiocytosis
- hypothalamic 158
- Hormone binding 212, 243
- Hypercapnia 131
- Hypernatremia 158
- Hypocapnia 131
- Hypodipsia 158
- Hyponatremia 141, 150, 162
- Hypotension 131, 132
- Hypothalamus
- electrical potential 78
  - nuclear groups 24
  - oxytocin 31
  - vasopressin 31
- Hypothalamic island 71, 72
- Hypothyroid 141
- Hypoxia 130–132
- Indomethacin 148
- Internuclear zone 67
- Intracerebroventricular infusion 119, 120
- Isoproterenol 145, 146
- Juxtaventricular sodium-sensitive mechanism 119
- Labour inducement 105
- Learning
- vasopressin 201, 202, 204–206
- Leucine aminopeptidase 174
- Leucine vasopressin 202
- Limbic system 72
- Lithium 164
- Lysine vasopressin 160, 201, 217
- Magnocellular neurons 22, 23, 59–62, 67–74, 79, 84
- Meclofenamate 148
- Memory 201, 203–206
- Microfilament 232
- Microtubule 220, 227
- Microvesicle 92
- Milk ejection 102, 105
- Mineralocorticoid 149
- Monoaminergic synapse 70
- Morphine 129, 206
- MSH 181, 201, 203
- MSH-RIF 181
- Muscles, striated 96
- Myxedema 140, 141
- Narcotic antagonists 162
- Natriuretic response 241
- Neurohypophysial hormones
- antidiuretic action 236
  - ATP 95
  - evolution of 3
  - female reproductive tract 6, 107, 108
  - papillary collecting duct 238, 239
  - physiological functions, theories
    - of release 6
    - calcium 95, 110
    - cyclic AMP 110

- Sodium depletion
  - angiotensin levels 123
- Sodium detector hypothesis 70, 119
- Sodium elevation
  - cerebrospinal fluid 119
- Sodium-potassium ATPase
  - inhibition 120
  - renal cortex 237
  - response to PTH 238
  - specific activity 238
- Sodium transport 95
- SQ 20,009
  - vasopressin release 111
- Stimulus-contraction coupling 96
- Stimulus-secretion coupling 96
- Suprachiasmatic nucleus 22, 26
- Supraoptic nucleus
  - action potential 78
  - afferent input 72
  - anesthetic effects 73
  - carotid occlusion effect 71
  - cholinergic synapse 67–74
  - functions of 80
  - inhibition 73
  - location 22, 24, 26, 27, 60
  - magnocellular neurons 67, 71–74
  - milk ejection 102, 105
  - nociceptory activation 72
  - orthodromic excitation 72
  - osmosensitive neurons 72
  - osmotic stimuli 71, 81–83
  - oxytocin 71, 74
  - recurrent inhibition pathway 70, 74
- Synaptic transmitters
  - drug actions 73, 74
- Syndrome of inappropriate ADH secretion 161, 164
  
- Theophylline
  - vasopressin release 111, 112
- 4-Threonine, 7-glycine oxytocin 16
- 4-Threonine-oxytocin 11–13
- Transport
  - secretory granules 96
  - transepithelial 243
  
- Vaginal dilation 107
- 4-Valine-arginine-vasopressin 11
- Vasopressin
  - adrenal insufficiency 139
  - behavior 201, 202, 204, 206, 207
  - binding 192
  - circadian rhythm 71, 72
  - clearance 189, 192, 193, 200
  - diffusion rate 192
  - effect of
    - catecholamines 146
    - prostaglandin 148
  - effect on
    - adenylate cyclase 211, 214–217
    - Henle's loop 211
    - renal medulla 240
  - hydroosmotic reactivity 236
  - learning 201, 202, 204–206
  - memory 201, 203–206
  - myxedema 140, 141
  - neurophysin 58, 60
  - neurosecretory granules 97, 98
  - norepinephrine 144–146
  - plasma concentrations 128, 212
  - potency 202, 217
  - receptor 212
  - release 59, 70–73, 78, 215
    - adrenergic step 131
    - anesthesia 73, 128, 129
    - angiotensin 110
    - atrial distention 146
    - behavior 71–73
    - blood gas tensions 128
    - blood pressure 128, 131, 132, 146
    - 8Br-cyclic AMP 111
    - caffeine 110
    - cyclic AMP 110, 111, 115
    - cyclic GMP 111
    - electrical stimulation 78
    - energy dependence 97
    - extracellular fluid volume 128, 150
    - hypercapnia 131
    - hypocapnia 131
    - hypotension 131, 132
    - hypoxia 131, 132
    - magnocellular neurons 67, 68, 71, 74
    - neurons, characterization of 80–82
    - neurons, firing frequency 79
    - nicotine 74
    - norepinephrine 147
    - osmotic influences 70, 73
    - pain 72
    - papaverine 111
    - phosphodiesterase 111

- potassium 111
- propagated action potential 88
- recurrent inhibitory pathway 74
- reflex pathway 71
- sleep-waking activity 71, 72
- SQ 20,009 111
- supraoptic nucleus 80
- theophylline 111
- tonic inhibition 131
- volume influences 71, 73
- structural changes 2, 11–18
- synthesis
  - magnocellular neurons 58–60, 64, 67
  - model 31–33
  - one cell-one hormone hypothesis 27
  - paraventricular nucleus 31
  - precursor protein 31, 33
  - prohormone 203
  - supraoptic nucleus 31
  - tubular secretion 192
  - treatment with 59, 60, 62
- Vasopressinase 174
- Vasopressin/oxytocin ratio 33, 38
- Water intake
  - renin-angiotensin system 125

