Journal of Neurochemistry
Raven Press, Ltd., New York
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Effect of Dexamethasone on Transport of α-Aminoisobutyric Acid and Sucrose Across the Blood–Brain Barrier

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Abstract: The effect of glucocorticoids on the blood–brain barrier (BBB) was studied in rats following a single injection or 3 days of dexamethasone administration. Tracers with a low permeability across the intact endothelium, [^{14}C]sucrose and α-[^{3}H]aminoisobutyric acid ([^{3}H]AIB), were simultaneously injected intravenously in untreated rats or in rats treated with dexamethasone. Unidirectional blood-to-brain transfer constants (K_i) in 14 regions of the rat brain were determined. In regions of control brain, average K_i values for AIB and sucrose were ~0.0020 and 0.00060 ml g^{-1} min^{-1}, respectively. The lowest transfer constants were found in caudate nucleus, hippocampus, white matter, and cerebellum. In dexamethasone-treated animals, K_i values for both sucrose and AIB markedly decreased by 30–50% in almost all brain regions. These results indicate that a single injection or 3 days of treatment with dexamethasone causes an apparent reduction in the normal BBB permeability, and dexamethasone may greatly interfere with drug delivery into brain. These observations may have an importance for the administration of drugs in brain disease in the presence of steroids.
Key Words: α-Aminoisobutyric acid—Blood–brain barrier—Dexamethasone—Corticosteroids—Cerebrovascular permeability. **Ziylan Y. Z. et al.** Effect of dexamethasone on transport of α-aminoisobutyric acid and sucrose across the blood–brain barrier. *J. Neurochem.* **51,** 1338–1342 (1988).

Anatomic and physiologic evidence indicates that an intact blood–brain barrier (BBB) is a prerequisite for maintaining homeostasis within the CNS. The BBB, a continuous cellular layer formed by endothelial cells of brain capillaries that are joined by tight junctions (zonulae occludens), restricts the passage of polar compounds and macromolecules from blood into the brain interstitium (Reese and Karnovsky, 1967; Rapoport, 1976; Bradbury, 1979). Several lipophilic drugs, some of which are widely used (local anesthetics, steroids, and tranquilizers), are known to alter membrane permeability (Seeman, 1966). Some of these drugs could influence BBB permeability. Steroids are of interest because dexamethasone (DXN) and other synthetic glucocorticoids are commonly used in clinical treatment for cerebral edema (Rasmussen and Gulati, 1962; Fishman, 1982).

Numerous studies have demonstrated that steroids can result in a rapid and dramatic decrease in the flux of water across the BBB (Fenske et al., 1979; Yamada et al., 1979; Sztriha et al., 1986). DXN also lowers experimentally induced increased cerebral vascular permeability associated with drug-induced hypertension (Bloomstrand et al., 1975; Johansson, 1978; Ziylan et al., 1984*a*), convulsive seizure activity (Eisenberg et al., 1970; Sztriha et al., 1986), ethanol toxicity (Rosengren and Persson, 1979), hyperammonemic coma (Sears et al., 1985), tumor (Shapiro and Posner, 1974; Yamada et al., 1979), cerebral infarction and hypoxia (Fenske et al., 1979; Barbrosa-Coutinho et al., 1985), and osmotic BBB injury (Neuwelt et al., 1982). It has also been shown that DXN reduces the normal permeability of cerebral blood vessel to horseradish peroxidase in mice (Hedley-Whyte and Hsu, 1986) and the permeability surface area product (*PA*) for water in the cerebral cortex of rats (Reid et al., 1983*a,b*).

These observations and the fact that both sympathetic stimulation (Edvinsson et al., 1977; Bill and Linder 1979) and corticosterone replacement after total adrenalectomy (Long and Holoday, 1985) are associated with a decrease in the permeability of the BBB to macromolecules led us to speculate that adrenal

Received November 24, 1987; revised manuscript received March 14, 1988; accepted April 29, 1988.
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Abbreviations used: ACTH, adrenocorticotrophic hormone; AIB, α-aminoisobutyric acid; BBB, blood–brain barrier; DXN, dexamethasone; K_i, unidirectional blood-to-brain transfer constant; *PA*, permeability surface area product.

corticosteroids might also specifically influence the permeability characteristics of the undisrupted normal brain microvasculature. Therefore, we investigated the effect of acute and subacute DXN administration on the permeability of the BBB by using isotopically labeled small compounds with a low permeability across the intact endothelium, such as sucrose (Ohno et al., 1978) and α-aminoisobutyric acid (AIB) (Blasberg et al., 1983).

MATERIALS AND METHODS

Twenty-six adult male Sprague-Dawley rats weighing 200–250 g were subjected to studies of the effect of DXN on BBB permeability. DXN (Soludecadron; Perouse Rantigny Laboratories, France) was administered at 2 mg kg^{-1} daily for 3 days or was injected intraperitoneally 3 h before the injection of radiotracers. A control group comprised six animals receiving the same amount of isotonic saline (0.9% wt/vol NaCl).

Animal preparation

The rats were anesthetized with pentobarbital sodium (35 mg kg^{-1} i.p.), and catheters filled with 100 IU of heparin in isotonic saline were inserted into a femoral vein and artery for blood sampling and administration of radiotracers. Body temperatures were monitored with a rectal thermometer probe, and external heat lamps were used to maintain body temperature at 35–37°C.

Isotopically labeled substances

[^{3}H]AIB (33.5 Ci/mmol) was obtained from New England Nuclear (Boston, MA, U.S.A.), and [U-^{14}C]sucrose (>350 mCi/mmol) and [^{3}H]dextran (140 mCi/g) were from Amersham International (U.K.). The molecular weights for [^{3}H]AIB, [^{14}C]sucrose, and [^{3}H]dextran were reported to be 103, 342, and 70,000, respectively, with 99% purity.

Experimental procedure

Cerebrovascular permeability was determined simultaneously for [^{3}H]AIB and [^{14}C]sucrose. Five microcuries of [^{14}C]sucrose and 25 μCi of [^{3}H]AIB were injected intravenously as a bolus in DXN-treated or in untreated control rats. Serial blood samples (100–150 μl/sample) were drawn from the femoral artery and rapidly centrifuged. The animals were decapitated at the end of the experiment, 10 min after injection of radiotracers. The brain was quickly removed and dissected into several regions. All tissue samples were placed in preweighed counting vials and reweighed to determine their weights. Sample solubilization was accomplished by adding 1 ml of Soluene-350 (Packard Instruments, Downers Grove, IL, U.S.A.). Whole blood samples were decolorized with hydrogen peroxide before counting. Finally, 10 ml of scintillation fluid {Toluene Scintilator; 0.1 g/L of 1,4-di[2-(5-phenyloxazolyl)]benzene and 5 g/L of 2,5-diphenyloxazole; Packard} was added to all tissue, plasma, and blood samples, and counting was carried out in an Intertechnique SL 3000 liquid scintillation spectrometer. All samples counts were approximately corrected for background and quenching.

Calculation

The unidirectional blood-to-brain transfer constant (K_i) was calculated in control and DXN-treated rat brains for [^{14}C]sucrose and [^{3}H]AIB from the tissue and plasma radioactivity data using the equation developed by Ohno et al. (1978). In brief, the initial rate for blood-to-brain transfer can be calculated by the following equation, when the passage of tracer into brain is assumed to be proportional to its plasma concentration and that the backflux (from brain to blood) is much smaller than the influx (from blood to brain) during the experimental period (T):

$$K_i = \frac{C_{br}}{\int_0^T C_{pl} \cdot dt} \quad (1)$$

where C_{br} is the parenchymal brain concentration of the tracer at the end of experiment (dpm g^{-1}), T is the duration of the experiment (min), and C_{pl} is the arterial plasma concentration (dpm ml^{-1}).

The transfer constant (K_i) is dependent on both the product of PA and the blood flow (F) (Renkin, 1959; Crone, 1963), namely,

$$PA = F \cdot \ln[1 - (K_i/F)] \quad (2)$$

With low permeabilities ($PA \ll F$), a condition well satisfied for sucrose (Ohno et al., 1978) and AIB (Blasberg et al., 1983), then, from Eq. 2, PA approaches K_i (Fenstermacher et al., 1981) and can be expressed in this measurement in term of a plasma clearance (ml g^{-1} min^{-1}).

The tissue parenchymal concentration (C_{br}) is obtained by subtracting the (dpm g^{-1}) value in the final whole blood and regional blood volume (ml g^{-1}) from the total regional brain concentration. Therefore, Eq. 1 may be written as

$$K_i = \frac{C_{br}(T) - VC_{wb}(T)}{\int_0^T C_{pl} \cdot dt} \quad (3)$$

where V is the regional blood volume and C_{wb} is the tracer concentration in the final whole blood volume.

Regional cerebral blood volume

The regional blood volume (V) was determined in control animals for the [^{3}H]dextran space, as the value for (dpm g^{-1} of brain/dpm ml^{-1} of whole blood) at the time of death. Rats were killed 1 min after intravenous injection of [^{3}H]dextran, when blood samples were collected. Radioactivity was determined in whole blood and brain regions as described above.

Statistical analysis

Student's t test for two independent groups was used to compare means of controls and DXN-treated animals. One-way analysis variance was used to compare the treatment groups, followed by Student's t test when the overall difference was statistically significant. Significance was taken as $p < 0.05$.

RESULTS

The K_i values calculated by Eq. 3 for [^{3}H]AIB and [^{14}C]sucrose in discrete brain regions of control and DXN-treated animals killed 10 min after intravenous injection of both radiotracers are shown in Table 1. The average values of K_i in brain regions of control rats for [^{3}H]AIB and [^{14}C]sucrose approximated 2.0 and 0.60 ml g^{-1} min^{-1} $\times 10^{-3}$, respectively. K_i values for [^{3}H]AIB in all brain regions were higher than similarly computed values for [^{14}C]sucrose and showed a high degree of correlation with the regional permeability pattern of sucrose, except in the hypothalamus.

TABLE 1. *Regional* K_i *values for [^{3}H]AIB and [^{14}C]sucrose in control and DXN-treated rats*

Brain region	[^{3}H]AIB Control	[^{3}H]AIB Single injection	[^{3}H]AIB 3-day treatment	[^{14}C]Sucrose Control	[^{14}C]Sucrose Single injection	[^{14}C]Sucrose 3-day treatment
	K_i (ml g^{-1} min^{-1} × 10^3)					
Olfactory bulb	3.16 ± 0.13	2.72 ± 0.12^a	1.91 ± 0.06^{a,b}	1.08 ± 0.08	1.01 ± 0.08	0.57 ± 0.02^{a,b}
Caudate nucleus	1.17 ± 0.06	0.83 ± 0.06^a	0.64 ± 0.03^{a,b}	0.46 ± 0.03	0.41 ± 0.03	0.29 ± 0.01^{a,b}
Hippocampus	1.74 ± 0.08	1.22 ± 0.07^a	1.02 ± 0.07^a	0.44 ± 0.03	0.37 ± 0.05	0.28 ± 0.02^a
Frontal lobe	2.51 ± 0.16	1.46 ± 0.05^a	1.40 ± 0.10^a	0.65 ± 0.06	0.42 ± 0.04^a	0.28 ± 0.01^{a,b}
Occipital lobe	2.13 ± 0.25	1.89 ± 0.10^a	1.32 ± 0.05^{a,b}	0.55 ± 0.05	0.50 ± 0.04	0.29 ± 0.03^{a,b}
Thalamus	1.68 ± 0.10	1.07 ± 0.07^a	1.01 ± 0.09^a	0.57 ± 0.05	0.52 ± 0.04	0.27 ± 0.01^{a,b}
Hypothalamus	4.17 ± 0.31	2.66 ± 0.18^a	2.10 ± 0.13^a	0.58 ± 0.04	0.55 ± 0.04	0.28 ± 0.01^{a,b}
Superior colliculus	2.01 ± 0.14	1.73 ± 0.19	1.03 ± 0.05^{a,b}	0.71 ± 0.05	0.63 ± 0.11	0.40 ± 0.03^a
Inferior colliculus	2.48 ± 0.18	1.91 ± 0.16^a	1.11 ± 0.06^{a,b}	0.72 ± 0.04	0.54 ± 0.11^a	0.52 ± 0.03^a
Cerebellum	2.00 ± 0.22	1.97 ± 0.10	1.23 ± 0.17^{a,b}	0.43 ± 0.02	0.40 ± 0.01	0.27 ± 0.01^{a,b}
Pons	2.52 ± 0.22	1.32 ± 0.66^a	1.32 ± 0.08^a	0.59 ± 0.05	0.61 ± 0.04	0.30 ± 0.01^{a,b}
Medulla	2.66 ± 0.14	2.02 ± 0.12^a	1.60 ± 0.10^{a,b}	0.67 ± 0.05	0.64 ± 0.01	0.37 ± 0.03^{a,b}
Midbrain	1.67 ± 0.10	1.44 ± 0.13	0.98 ± 0.04^{a,b}	0.55 ± 0.03	0.52 ± 0.04	0.29 ± 0.01^{a,b}
Gray matter	2.15 ± 0.20	1.85 ± 0.12	1.60 ± 0.06^{a,b}	0.58 ± 0.02	0.49 ± 0.04^a	0.28 ± 0.02^{a,b}
White matter (corpus callosum)	1.57 ± 0.10	1.30 ± 0.10	1.32 ± 0.10	0.48 ± 0.02	0.40 ± 0.05^a	0.37 ± 0.02^a

Data are mean ± SE values (n = 6–10). The K_i was calculated by Eq. 1 when the animals were killed 10 min after intravenous injection of DXN.

[a] Significant difference ($p < 0.05$) between control and DXN-treated animals.

[b] Significant difference ($p < 0.05$) between animals given a single injection and 3 days of DXN treatment.

The K_i for [^{3}H]AIB, but not for [^{14}C]sucrose, in the hypothalamus was markedly higher than in all the other brain regions. A lower transfer constant for both [^{3}H]AIB and [^{14}C]sucrose was found in caudate nucleus, hippocampus, and white matter.

A significant decrease in K_i, by 30–50%, for both [^{3}H]AIB and [^{14}C]sucrose was observed in almost all brain regions studied in rats pretreated for 3 days with DXN (Table 1); the K_i for both radiotracers in many brain regions was also reduced by ~20% in rats injected intraperitoneally with 2 mg kg^{-1}, 3 h before decapitation. The effect of a single injection of DXN was not significant in the hypothalamus, medulla, pons, and cerebellum. The cortex and thalamus were the areas most sensitive to the stabilizing effect of DXN. Figure 1 illustrates changes in K_i values for [^{3}H]AIB and [^{14}C]sucrose after DXN treatment in a representative brain region (frontal lobe).

DISCUSSION

Although the protective effect of DXN on altered BBB permeability has been extensively explored in various experimental models, the effectiveness of the drug on normal cerebrovascular endothelium is still a matter of conjecture. The present study clearly demonstrated that administration of DXN for 3 days or acute treatment with 2 mg kg^{-1} i.p. before killing leads to a reduction in normal cerebrovascular endothelium permeability for AIB and sucrose. This finding is consistent with the demonstration that acute or continuous DXN administration produces a significant decrease in the PA value for water in the cerebral cortex (Reid et al., 1983*a,b*). Our findings are also in accord with a previous study demonstrating that adrenalectomy produced a significant increase in BBB permeability to ^{125}I-labeled bovine serum albumin, with the magnitude of the increase ranging from 25 to 49% in discrete brain regions (Long and Holoday, 1985).

Furthermore, our results also showed that there is a high degree of correlation between the regional PA patterns of a nonelectrolyte (sucrose) and those of a small neutral amino acid (AIB). Both these test substances cross through the endothelial cells quite slowly and satisfy the conditions for measuring barrier permeability when expressed in terms of unidirectional transfer (K_i)

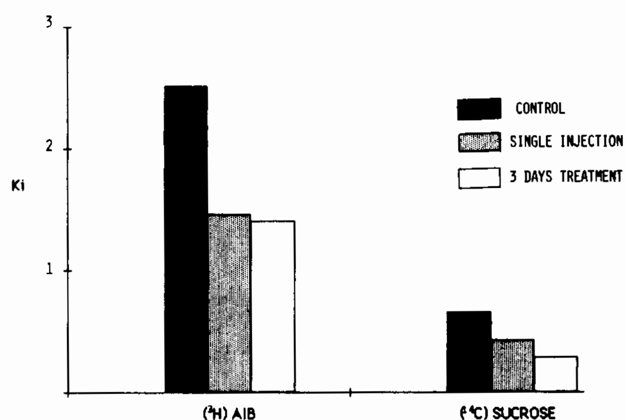

FIG. 1. Regional K_i values (in ml g^{-1} min^{-1} × 10^3) in a representative brain region (frontal lobe) for [^{3}H]AIB and [^{14}C]sucrose in rats pretreated with DXN for 3 days.

or *PA* (Ohno et al., 1978; Fenstermacher et al., 1981; Blasberg et al., 1983).

The regional K_i values for AIB in various brain regions of control animals are consistent with those found by Blasberg et al. (1983, 1984) and Ellison et al. (1986). The results for sucrose (both K_i and *PA* determinations) showed that K_i values as estimated by Eq. 3 are comparable with *PA* values and are also in agreement with cerebrovascular permeability measurements determined by the same equation in other experiments (Rapoport et al., 1980; Ziylan et al., 1983, 1984*b*).

Topographically, the effect of DXN on *PA* values for sucrose and AIB was regionally variable, as was shown briefly by Reid et al. (1983*a,b*) for water permeability. This result underscores the functional variety of different brain regions and may reflect variation in the local density of steroid receptors in the brain. It is in accord with studies demonstrating that the receptor system for glucocorticoids has a distinct neuroanatomical localization (McEwen, 1977; De Kloet, 1984).

The site and the mechanism of action of DXN on BBB permeability are not clearly established. DXN has multiple mechanisms of action, and the mode of this action on endothelial cell permeability in the normal brain and under pathological conditions may also differ. It participates in the pituitary-adrenal axis, which may regulate permeability of the BBB, either by a direct action or by a feedback influence on adrenocorticotrophic hormone (ACTH) (Long and Holoday, 1985); in the brain, DXN may modify aminergic or peptidergic system permeability (Ahmed et al., 1967; Raichle et al., 1975; Westergaard, 1975; McEwen, 1977; Rastogi and Singhal, 1978) or interact with other circulating substances, which then take part in the regulation of cerebrovascular permeability.

Alternatively, DXN may directly or indirectly participate in the cellular protein synthesis of vascular endothelium (Tosaki et al., 1985), which has been shown in nonneural tissues (Hirata et al., 1980).

The finding by Hedley-Whyte and Hsu (1986) that DXN treatment reduces the normal permeability of cerebral blood vessels to horseradish peroxidase by decreasing the number of horseradish peroxidase-containing small endothelial vesicles also suggests that DXN may have a direct effect on the vesicular transport.

Furthermore, the binding of labeled DXN primarily to the endothelial cells around the blood vessels (De Kloet, 1984), the finding by Rudman and Kutner (1978) that suppression of ACTH secretion or intercisternal administration of ACTH increases the penetration of labeled albumin, sucrose, insulin, or mannitol from blood into brain and CSF, and the relative effectiveness of intravenous ACTH on albumin penetration into brain suggest that glucocorticoids are more likely to affect the BBB through a direct action at the level of the vascular endothelium.

This reducing effect of glucocorticoids on cerebrovascular permeability, supported by the findings of Neuwelt et al. (1982), may also cause decreased drug delivery to the brain. Because DXN reduces the entry of macromolecules and diffusion-limited drugs, the therapeutic effect of adrenal cortical steroids, when steroids and other chemotherapeutic agents are combined in the treatment of CNS diseases, must be reevaluated.

REFERENCES

Ahmed A. B. J., George B. C., Gonzales-Auvert C., and Dingham J. R. (1967) Increased plasma arginine vasopressin in adrenocortical insufficiency and its inhibition by glucocorticoids. *J. Clin. Invest.* **47**, 111–123.

Barbrosa-Coutinho L. M., Hartman A., Hossmann K. A., and Ronnel T. (1985) Effect of dexamethasone on serum protein extravasation in experimental brain infarcts of monkey: an immunohistochemical study. *Acta Neuropathol. (Berl.)* **65**, 255–260.

Bill A. and Linder J. (1976) Sympathetic control of cerebral blood flow in acute arterial hypertension. *Acta Physiol. Scand.* **96**, 114–121.

Blasberg R. G., Fenstermacher J. O., and Patlak C. S. (1983) Transport of α-aminoisobutyric acid across brain capillary and cellular membranes. *J. Cereb. Blood Flow Metab.* **3**, 8–32.

Blasberg R., Molnar P., Groothuis D., Patlak C., Owers E., and Fenstermacher J. (1984) Concurrent measurements of blood flow and transcapillary transport in avian sarcoma virus-induced experimental brain tumors: implications for chemotherapy. *J. Pharmacol. Exp. Ther.* **231**, 724–735.

Bloomstrand C., Johansson B., and Rosengren B. (1975) Dexamethasone effect on blood–brain damage caused by acute hypertension in x-irradiated rabbits. *Acta Neurol. Scand.* **52**, 311–334.

Bradbury M. (1979) *The Concept of a Blood–Brain Barrier.* John Wiley and Sons, Chichester.

Crone C. (1963) Permeability of capillaries in various organs as determined by use of the indicator diffusion method. *Acta. Physiol. Scand.* **52**, 292–305.

De Kloet E. R. (1984) Function of steroid receptor system in the central nervous system. *Clin. Neuropharmacol.* **7**, 272–280.

Edvinsson L., Hardebo J. E., and Owmann C. (1977) Role of perivascular sympathetic nerves in autoregulation of cerebral blood flow and in blood–brain barrier functions. *Acta Neurol. Scand. [Suppl.]* **64**, 50–51.

Eisenberg H. M., Barlow C. F., and Lorenzo A. V. (1970) Effect of dexamethasone on altered brain vascular permeability. *Arch. Neurol.* **23**, 18–22.

Ellison M. D., Poulishock J. T., and Hayes R. L. (1986) Examination of the blood to brain transfer of α-aminoisobutyric acid and horseradish peroxidase. Regional alterations in blood–brain barrier function following acute hypertension. *J. Cereb. Blood Flow Metab.* **6**, 471–480.

Fenske A., Fisher M., Regli F., and Hase U. (1979) The response of focal ischemic cerebral edema to dexamethasone. *J. Neurol.* **220**, 199–209.

Fenstermacher J. D., Blasberg R. G., and Patlak C. S. (1981) Methods for quantifying the transport of drugs across brain barrier system. *Pharmacol. Ther.* **14**, 217–248.

Fishman R. A. (1982) Steroid in the treatment of brain edema. *N. Engl. J. Med.* **306**, 359–360.

Hedley-Whyte E. T. and Hsu D. W. (1986) Effect of dexamethasone on blood brain barrier in the normal mouse. *Ann. Neurol.* **19**, 373–377.

Hirata F., Schiffmann E., Venkatasubramanian K., Salamon D., and Axelved J. (1980) A phospholipase A_2 inhibitory protein in rabbit neutrophils induced by glucocorticoids. *Proc. Natl. Acad. Sci. USA* **77**, 2533–2536.

Johansson B. B. (1978) Effect of dexamethasone on protein extravasation in the brain in acute hypertension induced by amphetamine. *Acta Neurol. Scand.* **57**, 180–185.

Long J. B. and Holoday J. W. (1985) Blood–brain barrier: endogenous modulation by adrenal cortical function. *Science* **227**, 1580–1583.

McEwen B. S. (1977) Adrenal steroid feedback on neuroendocrine tissues. *Ann. NY Acad. Sci.* **297**, 568–579.

Neuwelt E. A., Barnet P. A., Bigner O. D., and Frenkel E. P. (1982) Effects of adrenocortical steroids and osmotic blood–brain barrier opening on methotrexate delivery to gliomas in the rodent: the factor of the blood–brain barrier. *Proc. Natl. Acad. Sci. USA* **79**, 4420–4423.

Ohno K., Pettigrew K. O., and Rapoport S. T. (1978) Lower limits of cerebrovascular permeability to nonelectrolytes in the conscious rat. *Am. J. Physiol.* **253**, H299–H307.

Raichle M. E., Hartmann B. K., Eichling J. O., and Sharpe L. G. (1975) Central noradrenergic regulation of cerebral blood flow and vascular permeability. *Proc. Natl. Acad. Sci. USA.* **72**, 3726–3730.

Rapoport S. I. (1976) *Blood–Brain Barrier in Physiology and Medicine.* Raven Press, New York.

Rapoport S. I., Fredericks W. R., Ohno K., and Pettigrew K. (1980) Quantitative aspects of reversible osmotic opening of the blood–brain barrier. *Am. J. Physiol.* **238**, R421–R431.

Rasmussen T. and Gulati D. R. (1962) Cortisone in the treatment of postoperative cerebral oedema. *J. Neurosurg.* **19**, 535–544.

Rastogi R. B. and Singhal R. L. (1978) Evidence for the role of adrenocortical hormones in the regulation of noradrenaline and dopamine metabolism in certain brain areas. *Br. J. Pharmacol.* **62**, 131–136.

Reese T. S. and Karnovsky M. J. (1967) Fine structural localization of a blood–brain barrier to exogenous peroxidase. *J. Cell Biol.* **34**, 207–217.

Reid A. C., Teasdale G. M., and McCulloch J. (1983*a*) The effect of dexamethasone administration and withdrawal on water permeability across the blood–brain barrier. *Ann. Neurol.* **13**, 28–31.

Reid A. C., Teasdale G. M., and McCulloch J. (1983*b*) Hormonal influence on water permeability across the blood–brain barrier. *Clin. Exp. Neurol.* **19**, 50–53.

Renkin E. M. (1959) Transport of potassium-42 from blood to tissue in isolated mammalian skeletal muscle. *Am. J. Physiol.* **197**, 1205–1210.

Rosengren L. E. and Persson L. I. (1979) Influence of ethanol and dexamethasone on blood–brain barrier dysfunction to albumin. *Acta Neurol. Scand.* **59**, 119–126.

Rudman D. and Kutner M. H. (1978) Melanotropic peptides increase permeability of plasma/cerebrospinal fluid. *Am. J. Physiol.* **234**, E327–E332.

Sears E. S., McCandles D. W., and Chandler M. D. (1985) Disruption of the blood–brain barrier in hyperammonemic coma and the pharmacologic effects of dexamethasone and difluoromethyl ornithine. *Neuroscience* **14**, 255–261.

Seeman P. M. (1966) Membrane stabilization by drugs: tranquilizers, steroids and anesthetics, in *International Review of Neurobiology, Vol. 9* (Pfeiffer C. C. and Smythies W. R., eds), pp. 145–221. Academic Press, New York.

Shapiro W. R. and Posner J: B. (1974) Corticosteroid hormones effects in an experimental brain tumor. *Arch. Neurol.* **30**, 217–221.

Sztriha L., Joó F., Szerdahelyi P., and Koltai M. (1986) Effect of dexamethasone on brain oedema induced by kainic acid. *Neuroscience* **17**, 107–114.

Tosaki A., Kolti M., Joó F., Adam G., Szerdahelyi P., Lepran I., and Szeteres L. (1985) Actinomycin D suppresses the protective effect of dexamethasone in rats affected by global cerebral ischemia. *Stroke* **16**, 501–505.

Westergaard E. (1975) Enhanced vesicular transport of exogenous peroxidase across cerebral vessels induced by serotonin. *Acta Neuropathol. (Berl.)* **32**, 27–42.

Yamada K., Bremer A. M., and West C. R. (1979) Effects of dexamethasone on tumor-induced brain oedema and its distribution in the brain of monkeys. *J. Neurosurg.* **50**, 361–367.

Ziylan Y. Z., Robinson P. J., and Rapoport S. I. (1983) Differential blood–brain barrier permeabilities to (^{14}C)sucrose and (^{3}H)inulin after osmotic opening. *Exp. Neurol.* **79**, 845–857.

Ziylan Y. Z., Agcaoglu G., and Gokhan N. (1984*a*) Effect of dexamethasone on the opening and reclosure time of the blood–brain barrier during acute drug-induced hypertension. *IRCS Med. Sci.* **12**, 1095–1096.

Ziylan Y. Z., Robinson P. J., and Rapoport S. I. (1984*b*) Blood–brain barrier permeability to sucrose and dextran after osmotic opening. *Am. J. Physiol.* **247**, R634–R638.