EDITORIAL REVIEW

Barrier Mechanisms for Neurotransmitter Monoamines and Their Precursors at the Blood-Brain Interface

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The integrity of the endothelial cell lining of the cerebrovascular bed constitutes a *morphological* blood-brain barrier mechanism to neurotransmitter monoamines. Circulating monoamines are prevented from entering the brain primarily at the luminal membrane of the endothelial lining. The small percentage of amines that may pass this membrane is deaminated within the endothelial cells and pericytes of brain microvessels (capillaries, venules, and small veins) and, in the case of large parenchymal and pial vessels, in the smooth muscle layers, where O-methylation also takes place. In the choroid plexus a corresponding deamination and O-methylation takes place in the epithelial cells. The presence of these enzymes constitutes a further, *enzymatic,* blood-brain barrier in the brain vessels for these monoamines. The monoamine precursors L-3,4-dihydroxyphenylalanine (L-dopa) and L-5-hydroxytryptophan readily pass from the luminal endothelial cell membrane but are trapped by another enzymatic barrier mechanism. Within the endothelial cells and pericytes of the microvasculature, these compounds are decarboxylated to their corresponding amines and then immediately deaminated. One clinical implication of these enzymatic barrier mechanisms is the use of decarboxylase and monoamine oxidase inhibitors as adjuncts to L-dopa treatment of Parkinson disease; these substances facilitate the entry of L-dopa into brain and thus increase the amount of dopamine available at receptor sites. A brief hypertensive or hypertonic stimulus can transiently open the blood-brain barrier through an effect on endothelial cell linings. High circulating concentrations of monoamines can also open the morphological barrier, but probably only indirectly by inducing an acute rise in systemic blood pressure. Once the barrier is open, systemically administered monoamines enter the brain parenchyma, where they can induce pronounced changes in cerebral blood flow and metabolism.

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The concept of a barrier between the blood and the brain parenchyma originates from observations by Ehrlich [21] a century ago that certain aniline dyes pass freely from the circulation into peripheral tissues but are excluded from the central nervous system. Since then, numerous other substances have been found to be more or less prevented from entering the brain. Studies to clarify the mechanisms underlying the barrier properties have, naturally, included attempts by various techniques to demonstrate a difference between brain and peripheral tissue microvessels, the major site of exchange in the circulatory system. It soon became clear that such differences do indeed exist. That intracerebral microvessels (capillaries, venules, and small veins) are unique precisely in terms of barrier properties was first demonstrated in the 1960s by Bertler et al [4, 5], who found by histofluorescence and chemical methods that the passage into brain of certain amine precursors, such as the amino acids 3,4-dihydroxyphenylalanine (dopa) and 5-hydroxytryptophan (5-HTP), is prevented by an enzymatic mechanism residing in the endothelium and pericytes of the microvessels. Evidence has been presented for several other enzymatic barriers at the blood-brain interface utilizing enzymes such as γ-aminobutyric acid transaminase [25] and monoamine oxidase (MAO) [4]. Shortly afterward it was shown that barrier properties of central and peripheral vessels also differ in morphological respects, i.e., in terms of ultrastructural features of the "sealing" between apposing endothelial cells, the paucity of transendothelial pinocytosis, and the absence of endothelial fenestrations [95, 111], all of which almost totally prevent passage of macromolecules across the endothelial lining of the brain vascular wall (for review, see [90]).

Normal functions of the brain depend upon

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adequate control of the levels of monoamine neurotransmitters and their precursors in the brain extracellular fluid compartment. Accordingly, the entry of these substances from the circulation must be strictly regulated, and this is accomplished by the blood-brain barrier (BBB). The morphological component of the BBB impedes to a great extent, although not totally [80], the passage of water-soluble and polar substances, such as the neurotransmitters, into brain parenchyma. The existence of an enzymatic barrier adds to effective regulation of the passage of highly active substances from the blood. The present review elucidates certain properties of the morphological and enzymatic BBB with regard to neurotransmitter monoamines and their precursors as well as conditions for bypassing these barriers experimentally, and discusses how barrier opening may affect functional variables such as cerebral blood flow (CBF) and metabolism.

Enzymatic Blood-Brain Barrier to Transmitter Monoamine Precursors

Tyrosine Hydroxylase

The amino acid tyrosine, the main precursor of the neurotransmitter catecholamines (Fig 1), is present in the circulation at a high concentration (about 10^{-4} M [1]). The influx from the circulation to the brain of radioactive tracer amounts of L-tyrosine is high [80, 81]. This amino acid shares an uptake site for neutral amino acids at the BBB in common with phenylalanine, methionine, histidine, cysteine, valine, isoleucine, leucine, tryptophan, threonine, and dopa [11, 12, 81, 85]. At the blood-brain interface these neutral amino acids are transported via a sodium-dependent "L-system" [14, 104]. This facilitated transport mediates equilibration across the cell membranes bidirectionally. Because the influx of amino acids to the brain is balanced in the steady state by an efflux of amino acids derived from proteolysis, the rate of net uptake across the barrier is considerably less than the overall transport rate [86].

Tyrosine hydroxylase (TOH) is present in brain microvessels as well as pial vessels and parenchymal arterioles [40]. In the pial and parenchymal vessels the enzyme is primarily localized to perivascular sympathetic nerves. So far there is no evidence that TOH activity in isolated fractions of microvessels reflects anything other than synthesis of catecholamines stored in the nerves contaminating the fractions [40]. In this context it is notable that direct innervation of the endothelial cells and pericytes of brain capillaries has recently been demonstrated by electron microscopy [94, 101]. If TOH represented an enzymatic blood-brain barrier for tyrosine, introduction of L-tyrosine into the brain circulation would cause formation of dopa—and possibly also dopamine (DA)—in the brain microvessel wall; however, this is not the case, as evidenced by fluorescence microscopy in combination with pharmacological inhibition of aromatic L-amino acid decarboxylase (AAD) and MAO (unpublished observations), an experimental model that is discussed in the following section. The presence of an amino acid transport system across the blood-brain interface, rather than a possible enzymatic barrier mechanism, is therefore probably the main limiting factor in determining the availability of tyrosine to the brain [86].

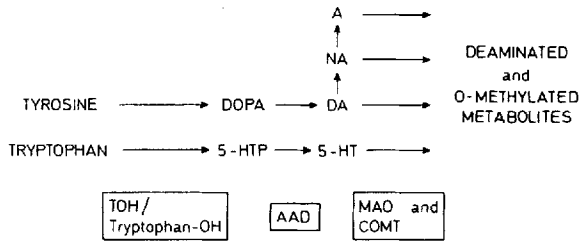

Fig 1. Enzymes related to the formation and degradation of neurotransmitter monoamines. (A = *adrenaline;* NA = *noradrenaline:* DOPA = *3,4-dihydroxyphenylalanine;* DA = *dopamine;* 5-HTP = *5-hydroxytryptophan;* 5-HT = *5-hydroxytryptamine;* TOH = *tyrosine hydroxylase;* AAD = *aromatic L-amino acid decarboxylase;* MAO = *monoamine oxidase;* COMT = *catechol-O-methyltransferase.)*

Aromatic L-Amino Acid Decarboxylase

The presence of AAD in the brain microvessel wall (Fig 2)—as distinct from large brain arteries, the vessels of the choroid plexuses, and peripheral vessels—was first shown in mice and rats by Bertler et al [4, 5] and Owman and Rosengren [84]. AAD activity was demonstrated histochemically and biochemically, as it was found that the amines DA and 5-hydroxytryptamine (5-HT) were formed within the microvessel wall subsequent to systemic administration of the corresponding substrates L-dopa and L-5-HTP. This finding has been confirmed by others in similar studies (e.g., [3, 16]) and also by measurement of AAD activity in isolated fractions of brain microvessels [35, 36, 40, 62, 99].

The circulating dopa concentration is only about 3×10^{-8} M [30], partly due to the efficient peripheral decarboxylation of dopa taking place in the gastrointestinal tract. 5-HTP still has not been detected normally in the circulation [72]. Because administration of L-dopa is of great clinical importance in the medical treatment of Parkinson disease, and this amino acid (as well as its amine product) can be visualized by histofluorescence techniques, it has been chosen as a model substrate for studying AAD activity even though it may not be the major sub-

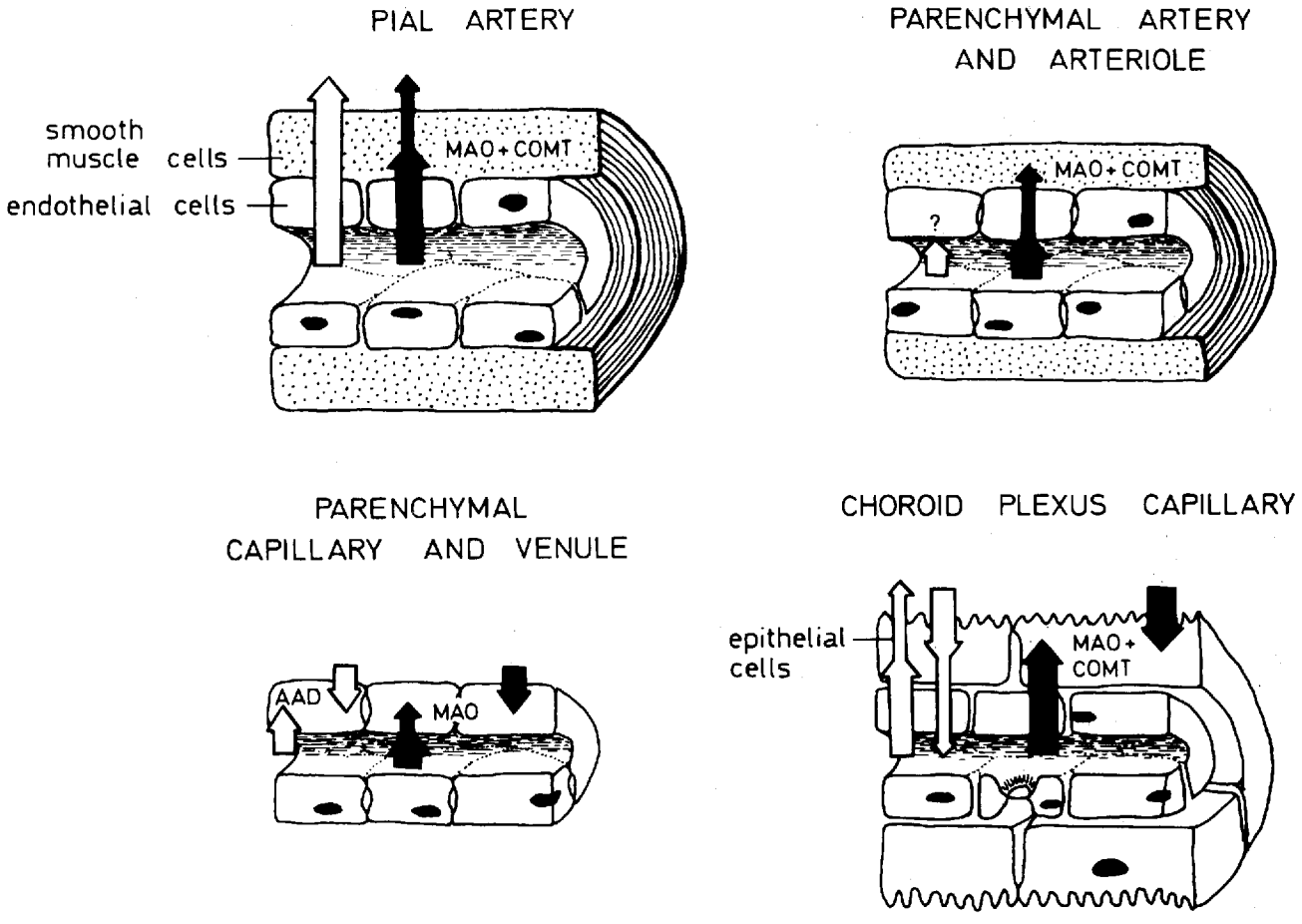

Fig 2. Morphological and enzymatic barrier mechanisms for neurotransmitter monoamines (filled arrows) *and their immediate precursors* (open arrows) *at various parts of the blood-brain interface. (Abbreviations the same as for Fig 1.)*

strate for microvascular AAD under physiological conditions.

L-Dopa and L-5-HTP are extracted to a high extent from the brain circulation [80, 105]. The uptake into brain has been characterized as a transport process occurring through facilitated diffusion [82, 104, 105] and, in addition, as an active, energy-dependent uptake process that has been found in the microvessels of various mammals including humans [41, 105]. The capacity of AAD to decarboxylate L-dopa in cerebral microvessels has an upper limit beyond which unchanged L-dopa leaks into the brain parenchyma. This has been estimated in rats by determination of the DA formed in two regions of the central nervous system devoid of DA neurons (cerebellum, and spinal cord caudal to chronic transection) following systemic L-dopa administration. In these regions, where synthesis of measurable amounts of DA can be expected to take place only in the microvessel walls, about 150 ng of DA is formed per gram of tissue per minute [39, 42]. The upper limit is reached at an intraperitoneal dose of about 100 mg per kilogram of body weight [39], corresponding to a circulating concentration of about 10^{-5} M [2]. The upper limit for the decarboxylation capacity has also been established by following the fate of systemically administered L-dopa using fluorescence histochemistry [17] or radiometric methods [105]. Decarboxylation in the brain microvessels is a rapid process; within less than half a minute, most of the amino acid injected has been converted to the corresponding amine [105].

Only slight regional differences in the rate of L-dopa decarboxylation at the level of the BBB have been found. Upon intracarotid injection of L-dopa, cortical regions have been shown to have a slightly lower rate of decarboxylation than deep hemispheric and brainstem regions [105]. Further, a slightly lower AAD activity was noticed in the cerebellum compared with the caudate nucleus and cerebral cortex. This was demonstrated in an in vivo model utilizing L-dopa administration in conjunction with decarboxylase inhibitor carbidopa, which is effective only peripherally and in brain microvascular walls, and by

determination of enzyme activity in isolated brain microvessels [40, 42]. On the other hand, great species differences in microvascular AAD activity exist: whereas considerable activity is found in humans, activity in the macaque monkey and in the baboon is very low, and the activities in rabbit and cat are also relatively low compared to other species [16, 17, 40, 41, 63]. Microvascular AAD activity is present in the fetal brain [41]. Peripheral microvessels revascularizing a transplant of cerebral tissue, as well as newly formed brain microvessels revascularizing a damaged brain area, are equipped with AAD activity [7, 100]. This is in contrast to the situation when brain microvessels regenerate into a peripheral transplant [100]. The findings indicate that the barrier characteristics of the newly formed microvessels are determined by properties of the tissue in which the new circulation occurs.

A minor amount of AAD activity is also found in preparations of pial vessels, large parenchymal vessels, and the choroid plexuses, but in these structures the enzyme is almost exclusively located in the perivascular sympathetic nerves [40, 67]. This is in contrast to the microvessels, in which the enzyme is doubtlessly located in the wall itself within the endothelial cells and pericytes [4, 40]. L-dopa uptake into the wall of pial vessels is very low (unpublished observations), supporting the conclusion that these vessels do not form part of the enzymatic barrier system of brain vessels. On the other hand, catechol-O-methyltransferase (COMT) is found in high concentrations in the pial vessel wall as well as in the choroid plexus [40, 67]. In this context it should be pointed out that most of administered L-dopa is metabolized to L-3-O-methyldopa and DOPAC before it reaches the central nervous system. It is possible that not only O-methylation but also transamination assists in impeding the passage of circulating L-dopa through pial vessels and the choroid plexuses.

One aspect of the role of AAD in BBB functions has been studied by measuring L-dopa–induced changes in CBF [22, 39, 48]: no marked cerebrovascular effects are obtained unless the enzymatic barrier is either inactivated by prior administration of the decarboxylase antagonist carbidopa or overloaded by high doses, or unless the morphological BBB has been opened. In these situations, increased CBF is probably a consequence of heightened cerebral metabolism mediated by stimulation of central dopaminergic receptors by the DA formed from L-dopa entering the brain parenchyma [48].

The efficiency of the BBB in impeding the passage of L-dopa into brain by a decarboxylating mechanism as well as the high AAD activity in peripheral tissues (represented by certain visceral organs and the sympathetic nervous system) have necessitated high doses of L-dopa in the medical treatment of Parkinson disease, with disturbing side effects as a frequent result. During treatment, circulating concentrations of up to about 10^{-5} M have been reported (e.g., [96]). Partly based on knowledge about the functions of the enzymatic BBB, it has been possible to overcome these problems to a considerable extent with addition of the peripheral decarboxylase inhibitors carbidopa or benserazide. These are effective in peripheral tissue as well as in brain microvessel walls, but only to a negligible extent in the brain parenchyma. Under these conditions the amine precursor can be administered in lower doses, thus reducing the incidence of undesired side effects (for review, see [87]).

Apart from metabolizing circulating L-dopa that would otherwise have been taken up across the BBB by the neutral amino acid transport system, the decarboxylase activity in brain microvessel walls may be involved in a more complex enzymatic barrier mechanism for other circulating amino acids such as phenylalanine, tyrosine, and tryptophan, or it may function as a link in a hitherto unknown metabolic pathway across the blood-brain interface.

Enzymatic Blood-Brain Barrier to Transmitter Monoamines

Monoamine Oxidase and Catechol-O-Methyltransferase

The normal circulating plasma levels of neurotransmitter monoamines in human beings is low: for norepinephrine the concentration is about 2×10^{-9} M [73, 77], and for 5-HT in platelet-free plasma about 5×10^{-8} M [98] unless its presence is due to inadvertent rupture of thrombocytes during handling. Circulating proteins bind about 50% of the normal plasma concentration of norepinephrine and a smaller fraction of 5-HT [15]. Because of the morphological BBB, only minor amounts (3 to 5%) of neurotransmitter monoamines are extracted from the brain circulation (see Fig 2), probably uniformly in various regions of the parenchyma [37, 80].

As already discussed, substantial amounts of monoamines may be formed indirectly in the microvessel walls due to local synthesis after uptake of the precursor amino acid. The amine formed is metabolized through the considerable MAO activity present in endothelial cells and pericytes of the brain microvessels [4, 35, 38, 40, 47, 61, 62, 99]. However, MAO activity also seems to play another role in the brain microvessels, namely, as a barrier for degradation of the minor amounts of circulating monoamines that may enter the cells of the microvessel walls. Naturally, monoamines other than the

well-known neurotransmitters norepinephrine, epinephrine, DA, and 5-HT may be substrates for this enzymatic barrier mechanism.

As mentioned, MAO as well as COMT activity is also found in pial vessels [39], choroid plexus [67], and parenchymal arterioles [40, 61, 99]. These enzymes are probably located in the smooth muscle cell layer of arteries, arterioles, and veins [66, 103]. In addition, MAO is also present in the perivascular sympathetic nerves. Vascular MAO of neuronal origin is usually found to be primarily of the A type, whereas MAO in the smooth muscle cell layer is mostly of type B [66]. In isolated brain vessels consisting of a mixture of large and small vessels, the presence of both A and B types of MAO has been reported even after removal of perivascular sympathetic nerves [61]. Hence, 5-HT, epinephrine, and norepinephrine (substrates primarily for type A) and DA (substrate to a varying degree for both type A and type B) may be deaminated within the walls of brain vessels [53, 113]. The primary action of these enzymes in arteries and arterioles may be related to local inactivation of vasoactive amines that reach the smooth muscle layer from perivascular nerves or from the circulation. In this latter respect, the uptake by smooth muscle [9, 26] and breakdown by MAO and COMT of amines that have penetrated beyond the endothelial lining may also represent a kind of barrier mechanism. This may be particularly important in the central nervous system, where passage across the endothelium is low and the subsequent degradation therefore can be expected to be efficient. That a small fraction of norepinephrine actually can reach the brain vascular smooth muscle cells and induce transient vasoconstriction before the amine is metabolized is consistent with the finding of a reduction in CBF, though only short lasting, after systemic or intracarotid injection of a bolus of norepinephrine.

Microvascular MAO, providing enzymatic breakdown of the amine trapped within endothelial cells and pericytes, should be considered not only as a barrier mechanism against the entrance of circulating neurotransmitters into the brain parenchyma, but also as one mode of inactivation of excess neurotransmitter present in the brain extracellular compartment, since monoamines are actively taken up into the wall of microvessels from the abluminal side [29, 38, 47]. During ischemia-anoxia, a transient increase in neurotransmitter monoamine levels occurs in the brain extracellular fluid compartment [77, 78, 112]. This excess, which may be detrimental to the brain by increasing metabolism in the ischemic-anoxic area [69], may reflect impaired reuptake of the transmitter not only by neurons and glial cells, but also by the local microvascular wall and the choroid plexuses. All these uptake mechanisms are considerably reduced by anoxia [29, 47, 67, 102]

It is possible that the brain microvascular endothelium with its MAO activity may serve more than a simple barrier function. The amine uptake [47] and enzymatic mechanisms may play a role in the function of endothelial cells themselves, which may be target structures for circulating and neurogenically released amines in, for example, contractile [83] and permeability [18, 89] processes. Direct innervation of the endothelial cells and pericytes of capillaries has recently been demonstrated by electron microscopy [94, 101].

The functional importance of the presence of MAO and COMT at the blood-brain interface has been elucidated by studies of the effect of amines on CBF and brain metabolism. The cerebrovascular response to circulating amines that have been administered systemically or locally into the internal carotid distribution can resemble, when MAO or COMT is inhibited [46, 70, 74, 75], the effects of amines administered intraventricularly, which thus bypass the BBB [70].

In vitro studies have indicated that human brain MAO is primarily of type B and that DA in human brain is a type B substrate [27, 113]. In the treatment of Parkinson disease, the MAO B inhibitor deprenil has recently been added to the combination of L-dopa and a peripheral decarboxylase inhibitor [6]. The therapeutic improvement resulting from this combination treatment is believed to be caused by an enhanced amount of DA available at DA receptor sites through inhibition of brain MAO. If the MAO present in human brain microvessels is also of the B type, an additional explanation would be inhibition of the enzymatic barrier mechanism to DA, thus reducing the loss of amine formed in the microvessel wall and brain parenchyma from the administered L-dopa.

It has been suggested that migraine sufferers have a defective MAO barrier [51]. This would lead to enhanced access to the brain parenchyma of, for example, norepinephrine, 5-HT, tyramine, and phenylethylamine, all of which have been implicated to varying degrees in the pathogenesis of a migraine attack.

Dopamine-β-hydroxylase

Dopamine-β-hydroxylase, which converts DA to norepinephrine and is a marker for norepinephrine innervation, is found in isolated fractions of large and small brain vessels [62, 99]. There is no evidence so far that dopamine-β-hydroxylase is located in endothelial cells of the brain vasculature; rather, its pre-

sence in this fraction reflects contamination with perivascular nerves. Its neuronal localization at the blood-brain interface cannot be expected to function as a barrier mechanism to DA.

Morphological Blood-Brain Barrier to Transmitter Monoamines

It can be assumed that an efficient blood-brain barrier to the monoamine neurotransmitters DA, norepinephrine, epinephrine, and 5-HT is necessary for adequate neurotransmitter function in the brain. Only in newborn animals, in which the morphological barrier is not yet fully developed, has substantial microvascular uptake of circulating neurotransmitter monoamines been demonstrated [68]. In adult animals, with a fully developed morphological BBB, fluorescence microscopy after systemic administration of DA has indicated that passage of circulating amines is greatly impeded at the luminal surface of the brain vessel [4, 38]. After systemic injection of norepinephrine or DA, amines have been found to leak into the vessel wall only at the level of large pial arteries [4, 38, 97]. These vessels, however, are probably not equipped with a morphological BBB (see discussion in [20, 38]).

As previously mentioned, extraction from the brain circulation of trace amounts of norepinephrine, epinephrine, DA, and 5-HT is on the order of 3 to 5% [37, 43, 80]. This explains why only minor passage of these amines into the brain parenchyma has been demonstrated after their systemic administration [10, 107, 108, 114]. Because of the presence of the previously discussed enzymatic barrier to these monoamines at the blood-brain interface, it can be assumed that the minor amount of amine leaving the brain circulation is efficiently trapped within the walls of the brain vascular tree. The negligible penetration of amines across an intact endothelial barrier may be the main reason why little or no effect on total CBF is seen after intravascular administration of these substances in concentrations not greatly exceeding their circulating levels at resting conditions (for review, see [20]; see also [46]).

Various attempts have been made to open the morphological BBB (for review, see [90]). In recent years, hypertonic (hyperosmolar) or hypertensive insult has been the most widely applied method of opening the BBB transiently. A pulse of hydrostatic pressure that elevates intracarotid pressure above 200 mm Hg has been shown to open the BBB in rats, as evidenced by extravasation of Evans blue–albumin complex [37, 45, 91]. Acute extreme systemic hypertension, induced by norepinephrine, epinephrine, metaraminol, or angiotensin, also causes BBB opening, probably related to pressure-forced overdistention of vessels [55, 59, 71]. In humans a corresponding acute rise in systemic pressure may lead to hypertensive encephalopathy, a syndrome to which hypertensive barrier opening probably contributes. BBB opening by the experimental approaches occurs preferentially in small arterioles and microvessels [37, 38, 54]. While the barrier opening is primarily confined to vessels in cortical structures of the hemisphere during the systemically induced hypertensive insult, a considerable barrier opening also occurs in deeper structures following a locally induced insult. Concomitant cerebral vasodilatation aggravates the BBB opening [34, 55, 58]. At the ultrastructural level, the barrier damage after systemically and locally induced acute hypertension is composed of channel formation in the cytoplasm of endothelial cells, increased transendothelial pinocytosis, and, rarely, opening of tight junctions between the cells [32, 33]. There is reason to believe that intracellular systems of microtubules are involved in this transendothelial transport since the extravasation caused by a hypertensive insult is counteracted by vincristine, an inhibitor of microtubular transport functions [64]. Reclosure of the barrier has been shown to occur within half an hour or less in these models [34, 57]. Also, intravascular administration of hypertonic solutions (e.g., urea) opens the BBB and allows large molecules that normally do not penetrate the barrier to pass from blood into brain tissue [8, 19, 38, 92]. The hypertonic barrier opening is reversible within a few hours [13, 33, 37, 88, 92].

It was originally suggested that hyperosmolar solutions extract intracellular water osmotically from cerebral endothelial cells, resulting in their shrinkage. Such shrinkage was believed to open the barrier transiently at the tight junctions between contiguous endothelial cells [8]. However, hyperosmolar solutions such as urea and mannitol have been shown to cause a considerable acute rise in systemic blood pressure [44], mainly via a central action [76]. Hyperosmolarity also has a direct vasodilatory effect [44, 60, 106]. Upon intracarotid infusion of a considerable volume of hyperosmolar solution, other factors may also contribute to barrier opening, such as a rise in intracarotid pressure, increased blood flow due to hemodilution, or ischemia [44]. The conclusion that opening of the BBB by a hypertonic solution is not due primarily to osmotic shrinkage of endothelial cells is also emphasized by the fact that blocking the increase in systemic blood pressure minimizes the barrier opening [33]. Furthermore, opened tight junctions have not actually been demonstrated ultrastructurally. Instead, increased transendothelial pinocytosis is seen [31], resembling findings during barrier opening induced by an acute hypertensive insult.

Following a hypertonic or hypertensive insult, the

brain uptake of various substances including norepinephrine can be enhanced several-fold in regions where the barrier has been opened [13, 33, 37, 56, 88, 93]. When the BBB is opened experimentally, an accumulation of monoamines in the cells of the microvessel wall is clearly distinguishable by fluorescence microscopy [23, 38]. Under these conditions, monoamines may enter the cytoplasm of endothelial cells by pinocytosis [32], or they may pass between or through the endothelial cells to reach the abluminal side of the endothelial membrane and from there enter endothelial cells as well as pericytes. An uptake process for monoamines into microvessel walls apparently works only across the abluminal membrane of the endothelial cell [47] in the direction from the brain into the cytoplasm of this cell (and of the pericyte). This would explain why monoamines accumulate in the microvessel walls in a narrow zone around the stitch channel and periventricularly when they are administered intraparenchymally [4] or into the ventricular system [24].

In contrast to the weak effect of neurotransmitter amines on CBF and brain metabolism when they are injected into the circulation of an animal with an intact BBB (for review, see [20]), the same compounds and concentrations induce substantial changes in these two functions following barrier opening by a hypertonic or hypertensive insult [19, 46, 50, 69] or when the barrier is circumvented through intraventricular administration of the amine [70]. It should be noted that the amine under study itself, when circulating in high concentrations, may induce a rise in blood pressure sufficiently pronounced to open the BBB. A clinical condition in which this may occur is the hypertensive crisis in patients with pheochromocytoma. This mechanism may also explain the pronounced increase in CBF and brain metabolism, probably mediated by circulating epinephrine, seen during immobilization stress in rats [11]. After the transmitter has penetrated the brain parenchyma, it activates amine-sensitive neurons, leading to a metabolically derived change in CBF. This effect of a given amine is not necessarily of the same kind as that obtained through its direct (but slight) action on the cerebrovascular wall from the lumen side when the BBB is intact.

When studying the degree of penetration achieved by circulating amines of high biological activity, the question arises whether the amine itself (besides its hypertensive effect) may influence permeability of the BBB. It has been reported that a minor opening of the morphological BBB may be obtained in the presence of high local concentrations of amines (e.g., norepinephrine and 5-HT). This barrier opening consists of enhanced vesicular transport across the cerebrovascular endothelial cells [109, 110]. However, the action of the amines does not seem to be a direct one because the changes fail to appear if their systemic effect on blood pressure is blocked [49]. Such high circulating concentrations may occur only during situations of extreme stress [11]. On the other hand, histamine and 5-HT, which are known to increase vascular permeability in peripheral vessels, may be released in high concentrations locally from mast cells present around cerebral vessels [18]. It should be emphasized that the effect of these amines on the morphological BBB has no bearing on quantitative measurements of their brain uptake [80], because in such studies only trace amounts of the amines are administered.

A further influence of catecholamines on BBB function has been demonstrated recently with the finding that stimulation of sympathetic nerves to the brain or of the locus ceruleus causes increased water permeability across the BBB [28, 89]. The importance of the small changes in water permeability induced by sympathetic stimulation is difficult to assess because the results imply that larger vessels also are involved in the water transport [65]. The effect of locus ceruleus stimulation is probably mediated through activation of β-adrenergic receptors recently shown to be present in brain microvessels [52, 79]. The observations suggest a function of neurogenic mechanisms and neurotransmitter monoamines at the blood-brain interface that may be of fundamental importance, especially in determining brain volume and osmolarity.

Concluding Remarks

Although it has been known for decades that a number of circulating substances are prevented from entering the brain, the principal nature of the BBB has recently become much better understood. It is now well established that the brain capillaries, in terms of barrier properties, differ both functionally and morphologically from capillaries in the peripheral circulation. Although most of the exchange between blood and tissue takes place at the capillary level, biologically highly active substances may also pass to some extent through the walls of the remainder of the cerebrovascular bed. These different types of vessels have a variety of components (endothelium, smooth muscle, nerves) that may contribute to barrier functions. In particular, the vascular endothelium is a highly complex metabolic organ—rather than a simple "lining" of the luminal surface of the vessel—designed to control the environment of the brain through a variety of mechanisms which we are only beginning to understand. Since the vessels of the choroid plexus are freely permeable, its epithelium constitutes an efficient trapping mechanism, because if substances had unlimited access to

the cerebrospinal fluid compartment they would easily reach the brain parenchyma.

The BBB is not, as previously thought, a static system established immutably during early life. A number of mechanical or pharmacological stimuli may open up the barrier more or less easily, depending on the efficiency of compensatory systems such as the sympathetic innervation of cerebral resistance vessels. Usually, reclosure takes place within minutes after the stimulus has subsided. However, it can be assumed that a protracted or permanent deficiency may be deleterious for an organ which has to control the entry of a variety of substances within narrow limits in order to maintain adequate metabolic and neurotransmitter functions.

Knowledge of the biochemical properties of the blood-brain interface has provided a basis for facilitating the entry of systemically administered drugs into the brain, a model example being the combined treatment of patients with Parkinson disease using L-dopa and enzyme inhibitors. It can be anticipated that similar approaches to overcome the barrier during medication will have wider applicability when the enzymatic features and transport mechanisms of the barrier have been elucidated in more detail.

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