Archives of Medical Research 45 (2014) 639−645

Archives of Medical Research

REVIEW ARTICLE

Glutamate Efflux at the Blood−Brain Barrier: Cellular Mechanisms and Potential Clinical Relevance

Helms Hans Christian Cederberg, Nielsen Carsten Uhd, and Birger Brodin

Department of Pharmacy, The Faculty of Health and Medical Sciences, University of Copenhagen, Universitetsparken 2, Copenhagen, Denmark

Received for publication October 30, 2014; accepted November 10, 2014 (ARCMED-D-14-00622).

L-Glutamate is considered the most important excitatory amino acid in the mammalian brain. Strict control of its concentration in the brain interstitial fluid is important to maintain neurotransmission and avoid excitotoxicity. The role of astrocytes in handling L-glutamate transport and metabolism is well known, however endothelial cells may also play an important role through mediating brain-to-blood L-glutamate efflux. Expression of excitatory amino acid transporters has been demonstrated in brain endothelial cells of bovine, human, murine, rat and porcine origin. These can account for high affinity concentrative uptake of L-glutamate from the brain interstitial fluid into the capillary endothelial cells. The mechanisms in between L-glutamate uptake in the endothelial cells and L-glutamate appearing in the blood are still unclear and may involve a luminal transporter for L-glutamate, metabolism of L-glutamate and transport of metabolites or a combination of the two. However, both *in vitro* and *in vivo* studies demonstrated blood-to-brain transport of L-glutamate, at least during pathological events. This review summarizes the current knowledge on the brain-to-blood L-glutamate efflux hypothesis including possible mechanisms to account for the transport, *in vivo* studies on blood glutamate scavenging and potential clinical relevance of the phenomenon. © 2014 IMSS. Published by Elsevier Inc.

Key Words: L-glutamate, Excitotoxicity, Excitatory amino acid transporters, Blood−brain barrier.

Introduction

L-glutamate is generally considered to be the most important excitatory neurotransmitter in the mammalian brain (1) but prolonged elevated concentrations of L-glutamate in the brain interstitial fluid (ISF) are highly cytotoxic, a phenomenon which has been termed excitotoxicity (2,3). Excitotoxicity occurs during a number of pathophysiological conditions such as traumatic brain injury, ischemic stroke, multiple sclerosis, epilepsy and Alzheimer's disease (4,5).

The regulation of L-glutamate in the brain ISF is dependent on glutamate transporters working in concert with intracellular metabolizing enzymes (6). The excitatory amino acid transporters, EAAT-1 (SLC1A3) and EAAT-2 (SLC1A2) are mainly localized in the plasma membrane of astrocytes (7−9) and have been shown to be essential for the control of extracellular L-glutamate levels (10,11). EAAT-3 (SLC1A1) is mainly localized in postsynaptic terminals in neurons (although astrocyte expression has also been demonstrated) and has less significant impact on the regulation of ISF L-glutamate levels (10,12). The uptake of L-glutamate via EAATs is indirectly dependent on the energy status of the cell because the translocation cycle includes co-transport of one molecule of L-glutamate with three sodium ions and one hydrogen ion and an exchange with one potassium ion (13,14). The transport activity and direction is thus coupled to the sodium-potassium ATPase (15).

Endothelial cells of the brain capillaries also express EAATs and may thus facilitate significant brain-to-blood L-glutamate efflux at least during pathological events (16−22). Previous reviews have focused on the role of EAATs in the brain endothelial cells and possible

Address reprint requests to: Birger Brodin, Group Leader, Associate Professor, Department of Pharmacy, The Faculty of Health and Medical Sciences, University of Copenhagen, Universitetsparken 2, Copenhagen, DK-2100, Denmark; Phone: +45 3530 61 69; FAX: +45 3530 6030; E-mail: birger.brodin@sund.ku.dk

0188-4409/$ - see front matter. Copyright © 2014 IMSS. Published by Elsevier Inc.
http://dx.doi.org/10.1016/j.arcmed.2014.11.004

blood-to-brain transport of L-glutamate originating from dietary monosodium-glutamate (23), on EAATs and blood L-glutamate scavenging (18) and on possible therapeutic applications of blood glutamate scavenging (16).

The present review aims to give an updated overview of mechanistic evidence on the molecular mechanisms of the blood–brain barrier (BBB) in the handling of brain L-glutamate, as well as a brief overview of animal studies related to lowering blood L-glutamate levels and brain ISF L-glutamate levels. Finally we will discuss the possible clinical relevance of brain glutamate efflux, and point to future directions of research.

Early In Vivo *Evidence for Efflux of L-glutamate from Brain Parenchyma to the Blood*

L-glutamate concentration in plasma is in the range of 30–90 μM and varies depending on nutritional status and exercise (24,25). L-glutamate is accumulated in erythrocytes, where the concentration has been estimated to range from 280 μM (24) to 450 μM (26). The mean blood concentration of L-glutamate will thus be an average of plasma and erythrocyte L-glutamate concentrations, corrected for their volume fractions, estimated to be ~140 μM (24).

The concentration of L-glutamate in the ISF of the resting, undisturbed brain parenchyma is not readily measured but estimates point at concentrations in the lower micromolar/submicromolar range. Values from 0.1–3 μM have been recorded, using microdialysis techniques (27). The large concentration difference between plasma and brain extracellular fluid indicates that the BBB has a very low permeability for L-glutamate, an active efflux of L-glutamate, a rapid brain metabolism of L-glutamate or a combination of these. The uptake of L-glutamate from blood to brain is low (28,29) and has been hypothesized to take place via a saturable carrier mechanism (30). However, brain microvessel concentrations of L-glutamate has been estimated to ~750 nmol g^{-1} (31), equivalent to ~785 μM assuming that endothelial cells have a density 1.048 g/mL (32) and that the L-glutamate is exclusively present in endothelial cells and not astrocyte or neuron remnants contaminating the microvessels. If this estimate is correct, then the L-glutamate concentration gradient will be unfavorable for uptake from the blood and into the endothelial cells. However, there would be a measurable unidirectional isotope uptake flux as observed by Oldendorf and Szabo, but not a net uptake (30). Efflux of L-glutamate from the brain was demonstrated by Drewes and colleagues (33) in studies on perfused dog brains. Concentrations of amino acids were measured in the arterial and venous perfusates and net movements of amino acids were calculated from the differences, indicating a net efflux of L-glutamate from the brain in the order of ~1 μmol 100 g $brain^{-1}$ min^{-1} under basal conditions (33). Based on these data, Pardrigde suggested that an active L-glutamate efflux system must be present at the BBB (34). This suggestion was supported by Hutchison and colleagues who investigated uptake kinetics of L-glutamate into isolated rat brain capillaries (35). They demonstrated that a high affinity (K_M ~2 μM), temperature dependent and ouabain sensitive L-glutamate uptake system was present in the capillaries, which could account for the previous observed brain efflux. This was further supported by studies by Hosoya et al. who performed intracerebral microinjections of radioactive L-glutamate, L-aspartate and D-aspartate in rats (36). They demonstrated rapid clearance from the brain of L-glutamate and L-aspartate, which correlated with the appearance of the two isotopes in jugular vein samples, whereas D-aspartate stayed in the brain compartment. Hosoya et al. used thin layer chromatography to verify that at least the main part of the appearing radioactivity in the blood originated from intact L-aspartate/L-glutamate (~70 and 84% respectively) (36). Initially, this efflux was not believed to be associated with EAAT transporters, partly because of lack of D-aspartate efflux and partly because immunolabeling studies had not shown EAAT-1 and -2 expression in rat brain endothelial cells (7,8,36). However, in recent decades, evidence has accumulated that EAATs are present in brain capillary endothelial cells and may take part in the brain L-glutamate efflux (see sections below).

Studies of EAAT Expression and Activity

Table 1 provides an overview of reported EAAT subtype expression patterns in different preparations from different species.

Expression in Intact Brain Capillaries

Brain capillaries from mice have been shown to have a high expression of EAAT-3 mRNA (22,37,38) as well as protein expression of EAAT-1, -2 and -3 (although mainly subtypes -1 and -2) (39). Freshly isolated bovine brain capillaries express EAAT-1, -2 and -3 mRNA, whereas only EAAT-1 has been detected at the protein level (21,40). Large quantities of EAAT-1 mRNA (41) and protein (42) were found in human brain capillaries. These studies indicate that EAAT's are present in endothelial cells. However, contamination by glial tissue or remnants of astrocyte endfeet may have

Table 1. Overview of EAAT subtype expression patterns in different species

	EAAT-1	EAAT-2	EAAT-3	References
Capillaries or intact tissue preparations				
mRNA	B,H, M	B, M	M	(21,22,37,38,40,41)
Protein	B, H, M	M	M	(17,21,39,40,42)
Cell culture				
mRNA	B		B, M	(21,22)
Protein	B, M, P, R	M, P	B, M, P, R	(19–21,39)

B, bovine; H, human; M, mouse; P, porcine; R, rat.

Download English Version:

https://daneshyari.com/en/article/3446382

Download Persian Version:

https://daneshyari.com/article/3446382

Daneshyari.com