*Neurosciences à Nouzilly – 20ème Journée & Réunion annuelle de la SEISC*

# La barrière hémato-encéphalique dans tous ses états

Congrès organisé avec le sponsoring de CORNING

16 & 17 novembre 2017

Amphi A, site des Tanneurs, Université François Rabelais de Tours
3 Rue des Tanneurs, 37041 Tours, France

**Jeudi 16 novembre 2017**

**13h30** Accueil des participants

**14h00** **Introduction**
Yves Tillet, *FED4226*, UMR *Physiologie de la Reproduction et des Comportements, Centre INRA Val de Loire, Nouzilly &* Jean-François Ghersi-Egea, *Président de la SEISC*

**14h30** **The neuroprotective functions of blood-brain interfaces during development**
*Jean-François Ghersi-Egea, Inserm U1028, CNRL, Lyon*

**15h15** **Imagerie TEP pour l'étude des propriétés fonctionnelles de la BHE : implications neuropharmacocinétiques.**
*Nicolas Tournier, CEA SHFJ Saclay*

**16h00** Pause-café et Session Posters

**17h00** **Les tanycytes et la BHE**
Bénédicte Dehouck, Centre de Recherche Jean Pierre Aubert, UMR-S1172- Université Lille, CHRU, *Développement et plasticité du cerveau neuroendocrine Lille*

**17h45** **Role of ABCA7 in cellular cholesterol homeostasis and Aβ peptide efflux at the blood brain barrier level: implications in Alzheimer's disease**
Yordenca Lamartinière, *LBHE EA 2465, Lens*

**18h05** **The LXR/RXR axis and the blood-brain barrier: from cholesterol homeostasis to transport of amyloid-ß peptides by brain pericytes and brain capillary endothelial cells**
Julien Saint-Pol, *Université d'Artois EA 2465, Laboratoire de la Barrière Hémato-Encéphalique (LBHE), Lens*

## Vendredi 17 novembre 2017

**9h00** **Modern methods for delivery of drugs across the blood-brain barrier**
Jean-Michel Escoffre, *Inserm U930, Tours*

**9h45** **Modélisation de la barrière hémato-encéphalique**
Guylène Page, *CiMoThéMA, EA3808, Poitiers*

**10h30** Pause-café et Session Posters

**11h30** **Pediatric high-grade glioma: modelisation of the blood-tumor barrier using a human syngenic approach**
Clémence Deligne, *Université d'Artois EA 2465, Laboratoire de la Barrière Hémato-Encéphalique (LBHE), INSERM U908, Lens*

**11h50** **Protection of the cerebrospinal fluid by choroidal glutathione peroxidases during perinatal development**
Elodie Saudrais, *Fluid & BIP Facility, CRNL, Inserm U1028, CNRS UMR5292, Lyon*

**12 h10** **Effet de corps cétoniques sur la barrière hémato-encéphalique dans le cadre de la Maladie d'Alzheimer**
Pietra Candela, *Université d'Artois, EA2465, Laboratoire de la Barrière Hémato-Encéphalique (LBHE), Lens*

**12h30** Déjeuner

**14h00** **Modulation saisonnière de la perméabilité de la BHE chez un modèle expérimental, la brebis.**
Laurence Dufourny, UMR *Physiologie de la Reproduction et des Comportements, INRA-CNRS Univ. Tours IFCE, Centre INRA Val de Loire, Nouzilly*

**14h45** Assemblée Générale de la SEISC – Remise du prix

## Communications affichées

**Prediction of human clinical data based on human primary innovative in vitro models. Part I: The BBB Modelling**
Fabienne Glacial, Nicolas Perrière, *BrainPlotting, IPEPS-ICM, 47 Boulevard de l'Hopital, 75013 Paris*

**Integrity of blood-brain interfaces in an animal model of severe neonatal jaundice**
Blondel S, Guy R, Saudrais E, Gazzin S, Ghersi-Egea JF, Strazielle N, *Brain Interfaces Platform, CRNL, INSERM U1028-CNRS UMR5292, Lyon, FR*

**Lithium transport at the blood-brain barrier in in vivo rat and in vitro human models**
Huilong Luo, Matthieu Gauthier, Xi Tan, Christophe Landry, Joël Poupon, Marie-Pierre Dehouck, Fabien Gosselet, Nicolas Perrière, Frank Bellivier, Salvatore Cisternino, Xavier Declèves, *Inserm UMRS1144*

**Connexin 30 is expressed in a subtype of mouse brain pericytes**

Noémie Mazaré, Alice Gilbert, Anne-Cécile Boulay, Nathalie Rouach and Martine Cohen-Salmon, *CIRB, Collège de France, CNRS UMR 7241/INSERM U1050*

**Impact of three months ketogenic diet on brain microvessels of TgCRND8 mice**

Corsi Mariangela, Versele Romain, Fuso Andrea, Sevin Emmanuel, Di Lorenzo Cherubino, Businaro Rita, Fenart Laurence, Gosselet Fabien, Candela Pietra, *Univ. Artois, EA 2465, Laboratoire de la Barrière Hémato-Encéphalique (LBHE), F-62300 Lens,France*

## Résumés

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### The neuroprotective functions of blood-brain interfaces during development

*Jean-François Ghersi-Egea, Equipe FLUID et plateforme BIP, Centre de Recherche en Neurosciences de Lyon, INSERM U1028, CNRS UMR 5292, Université Claude Bernard Lyon-1*

The main blood-brain interfaces protecting the central nervous system comprise the blood-brain barrier located at the endothelium of the cerebral microvessels, and the blood-CSF barrier located at the epithelium of the choroid plexuses. Both endothelial and epithelial barrier cells display a tight phenotype very early on during fetal development. The brain grows and matures in a controlled environment provided by these interfaces. The influence of the choroidal barrier on the brain fluid environment and brain functions is especially relevant during development. The blood-brain interfaces display developmental stage-specific transport and metabolic properties that explain differences in blood-brain molecular exchanges between fetuses, infants and adults. Postnatal windows of blood-brain interface sensitivity to perinatal insults have been shown, that shape the neuroimmune response in the developing individual.

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### Imagerie TEP pour l'étude des propriétés fonctionnelles de la BHE : implications neuropharmacocinétiques.

*Nicolas Tournier, Imagerie Moléculaire In Vivo, IMIV, CEA, Inserm, CNRS, Univ. Paris-Sud, Université Paris Saclay, CEA-SHFJ, Orsay, France*

La tomographie par émission de positons (TEP) est une modalité d'imagerie dynamique et quantitative qui repose sur l'utilisation de radioligands spécifiques d'une cible donnée. Au niveau cérébral, la modélisation pharmacocinétique des données d'imagerie permet d'estimer la densité d'une cible pharmacologique mais également d'étudier le passage de composés à travers la barrière hémato-encéphalique (BHE) de manière non-invasive.

Ces dernières années, plusieurs équipes ont utilisé l'imagerie TEP pour mettre en évidence le rôle déterminant de systèmes de transport membranaires sur le passage cérébral de médicaments chez l'animal comme chez l'Homme. Aujourd'hui l'imagerie TEP représente une approche translationnelle pertinente pour l'évaluation de stratégies de contrôle ou d'optimisation du passage cérébral de composés d'intérêt.

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### Les tanycytes et la BHE

*Bénédicte Dehouck, Centre de recherche Jean Pierre Aubert, UMR-S1172- Université Lille - CHRU Développement et plasticité du cerveau neuroendocrine*

Les tanycytes sont des cellules ependymogliales que l'on trouve spécifiquement dans les organes circumventriculaires. Ces cellules forment une interface entre le sang et le liquide céphalorachidien. A l'instar de la barrière hémato-encéphalique, cette interface favorise les échanges entre le sang et le système nerveux central tout en maintenant l'homéostasie cérébrale. Les tanycytes ont été principalement étudiés au niveau de l'éminence médiane, l'organe circumventriculaire formant le plancher du troisième ventricule et matérialisant la jonction entre l'hypothalamus et la tige pituitaire de l'hypophyse. Les tanycytes de l'éminence médiane permettent la régulation de la sécrétion de neuropeptides tels que la GnRH et la TRH dans le sang porte hypohysaire ; mais ils sont aussi impliqués dans le transport des signaux hormonaux au noyau arqué hypothalamique, impliqué dans l'homéostasie énergétique. La plasticité des tanycytes et de l'interface qu'ils forment avec les vaisseaux cérébraux permet une régulation constante mais aussi adaptative de ces échanges bidirectionnels. L'altération des fonctions des tanycytes et de la plasticité de cette interface pourrait être à l'origine de pathologies métaboliques telles que l'obésité.

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## Modern methods for delivery of drugs across the blood-brain barrier

*Jean-Michel Escoffre,* *Inserm U930, Tours*

Due to an aging population and the occidental lifestyle, the treatment of brain diseases such as neurodegenerative diseases and brain cancer is a major public health issue. Nowadays, our country has more than one million people suffering from brain diseases. Despite the development of therapeutic molecules targeting the proteins involved in these diseases, the marketing of these molecules is limited by the lack of specific delivery of these molecules in the brain. Moreover, the inability of these molecules to cross the blood-brain barrier (BBB) is another major limitation. Current methods of intracerebral delivery of therapeutic molecules induce disturbance of the BBB throughout the volume of perfused tissue and can not be used for targeted delivery. In this context, the notion of active delivery is of great interest because it implies both effective formulation and better targeting. The specific delivery of drugs to the brain continues to be a major challenge for the treatment of brain diseases. In this conference, I invite you to discover the moderner methods for delivery of drugs across the blood-brain barrier.

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## Modélisation de la barrière hémato-encéphalique

*Guylène Page, EA3808 « Cibles moléculaires et Thérapeutique de la maladie d'Alzheimer », PBS, 1 rue Georges Bonnet, Bâtiment B36,1 Rue Georges Bonnet, Poitiers (http://cimothema.labo.univ-poitiers.fr)*

La barrière hémato-encéphalique est complexe non seulement au niveau structural par les nombreux acteurs cellulaires qui la composent mais aussi au niveau moléculaire par les multiples acteurs impliqués dans l'homéostasie qui règne autour d'elle mais aussi dans son dynamisme. Cette barrière par les nombreuses interactions cellulaires et moléculaires qui peuvent se mettre à l'interface du sang et du cerveau, constitue une véritable unité neurovasculaire dont l'idéal en recherche est de pouvoir la modéliser le mieux possible pour analyser correctement les questions posées au niveau de cet environnement. La conférence portera donc sur une revue de la littérature des modèles de la BHE actuellement disponibles tout en glissant les paramètres physiques d'étanchéité, la fonctionnalité et la sélectivité obtenues avec ces modèles. Entre les lignées et les cultures primaires, le savoir-faire est précieux. Il sera aussi intéressant d'aborder le développement de ces modèles en tant que produits commerciaux. A l'heure d'aujourd'hui, où les causes de nombreuses maladies neurodégénératives sont inconnues, l'unité neurovasculaire correctement reconstituée pourrait livrer ses secrets.

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## Modulation saisonnière de la perméabilité de la BHE chez un modèle expérimental, la brebis.

*Laurence Dufourny,* *UMR Physiologie de la Reproduction et des Comportements, INRA-CNRS-Univ. Tours-IFCE, Centre INRA Val de Loire, 37380 Nouzilly*

La durée du jour (photopériode) est le principal synchronisateur de l'activité saisonnière de reproduction chez plusieurs espèces de rente telles que les ovins, les caprins et les équins. Chez les ovins, principal modèle expérimental utilisé dans notre équipe, les jours courts (8H de lumière par 24H) permettent le déclenchement de la saison de reproduction alors que les jours longs provoquent un passage en repos sexuel. L'avantage de ce modèle expérimental est de permettre des approches de neurochirurgie stéréotaxique avec la mise en place de canules dans les ventricules cérébraux, celles-ci permettant le prélèvement de liquide céphalo-rachidien (LCR) sur une période de temps étendue et donc l'obtention de volumes conséquents de LCR. Les travaux réalisés depuis une quinzaine d'année ont permis d'étudier la modulation du passage des molécules de la périphérie vers le système nerveux central notamment les stéroïdes et les protéines, de mesurer la production de LCR selon la photopériode et d'essayer d'analyser les mécanismes impliqués au niveau des plexus choroïdes. Enfin dernièrement, une collaboration nous a permis d'élargir nos résultats aux perturbateurs endocriniens. Ces différents aspects seront documentés au cours de l'exposé.

COMMUNICATIONS ORALES

## The LXR/RXR axis and the blood-brain barrier: from cholesterol homeostasis to transport of amyloid-ß peptides by brain pericytes and brain capillary endothelial cells.

Julien Saint-Pol, *Laboratoire de la Barrière Hémato-Encéphalique (LBHE), EA2465, Université d'Artois, LENS, France*.

The understanding of the pathologic mechanisms of dementia and particularly Alzheimer's disease (AD) is a crucial medical challenge since 47 million people are affected worldwide. This pathology is in part associated with a lack of elimination and therefore an accumulation of amyloid-ß (Aß) peptides within the brain, neurovascular spaces and around brain microvessels forming the blood-brain barrier (BBB). BBB controls the bidirectional transport of Aß peptides, and this transport is altered in AD. Since the identification of Apoε4 allele in 1993 and then of single nucleotide polymorphisms (SNPs) in genes related to cholesterol homeostasis such as Abca7 in AD patients, a link between brain cholesterol homeostasis and the progression of AD was established. Cholesterol homeostasis is controlled by the Liver X nuclear Receptors (LXRs) which work in an obligatory heterodimer with Retinoid X Receptors (RXRs). Some studies have described the benefit effects of the activation of LXRs (by their natural endogenous cholesterol-derivated agonists referred to as oxysterols) and of RXRs (by bexarotene, a drug used in T-cell lymphoma treatment) in AD mice brains and perivascular spaces, but few of them focused at the BBB level. This presentation makes an overview of our recent data highlighting the importance of the BBB in the LXR/RXR-mediated effects in brain cholesterol metabolism and Aß peptide clearance and therefore in AD.

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## Role of ABCA7 in cellular cholesterol homeostasis and Aβ peptide efflux at the blood brain barrier level: implications in Alzheimer's disease

Yordenca LAMARTINIERE, LBHE EA 2465*, Université d'Artois, LENS, France*.

Because of its crucial role in the regulation of exchanges of molecules as beta amyloid peptides (Aβ) and cholesterol between blood and brain, blood brain barrier (BBB) plays a key role in neurodegenerative diseases such as Alzheimer's disease (AD). Recently, several genome wide association studies identified another genetic risk factor for AD, ABCA7 which loss of function mutations in the gene are closely linked to excessive accumulation of Aβ peptides and disturbed cholesterol homeostasis. We thus investigated the role of ABCA7 in cholesterol and Aβ peptides exchanges at the BBB level. In order to decipher the functions of this transporter at the brain vasculature level, we analyzed brain capillaries of Abca7 -/- mice and elaborated a mouse in vitro BBB model with a downregulation of Abca7 expression using small RNA interference technique (siABCA7 cells). Our results show that the downregulation of Abca7 expression is associated with a decrease of ABCA1 and ApoE expression as well as a reduction of cholesterol efflux from endothelial cells. We also observed a reduction in Aβ peptides basolateral-to-apical transport in the presence of ApoA-I, in siABCA7 cells. Thus this

data highlighted a role for the transporter ABCA7 in cholesterol homeostasis and amyloid clearance at the BBB level. Therefore, ABCA7 represents promising therapeutic target for AD.

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### Pediatric high grade glioma: modelisation of the blood-tumor barrier using a human syngenic approach

Clémence Deligne, *Laboratoire de la Barrière Hémato-Encéphalique (EA 2465), Université d'Artois, Faculté Jean Perrin, Lens, F-62300, France & Unité Tumorigénèse et Résistance aux Traitements (Inserm U908), Centre Oscar Lambret, Lille, F-59000, France*

Brain tumors are the most frequent solid tumors in children. Among them, diffuse midline glioma (DMG) is almost uniformly fatal and represents the leading cause of brain tumor-related death. One reason for the clinical failure is the poor access of chemotherapeutic agents to the brain parenchyma due to the presence of the blood-brain barrier (BBB). The BBB, located at the brain capillary endothelial cells, tightly controls the exchanges of molecules between the blood and the brain. In most pathological conditions, the BBB specific properties are modified. Thus, our aim is to better understand the influence of DMG on the physical and metabolic properties of the BBB, using an in vitro approach. Our model is based on the coculture of human endothelial cells differentiated from CD34+ stem cells with bovine pericytes (BLECs: Brain-Like Endothelial Cells). To adopt a syngenic approach, we characterized human pericytes and confirmed their ability to induce the BBB phenotype in our endothelial cells. This novel human syngenic model was validated in a new configuration, allowing the development of a triculture with human DMG cells. Our preliminary results revealed a physically intact BBB in the presence of DMG cells, which is consistent with clinical observation. In the long run, we will use this human blood-tumor barrier model to further investigate the influence of DMG on the BBB physiology and their consequent impact on the chemotherapeutic transport through the BBB, which could modify the tumor exposure to chemotherapeutic agents and consequently modulate treatment efficiency.

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### Protection of the cerebrospinal fluid by choroidal glutathione peroxidases during perinatal development

Elodie Saudrais, *Eq. Fluid & BIP Facility, CRNL, INSERM U1028, CNRS UMR5292, Lyon, France*

The developing brain is highly sensitive to oxidative stress and lipid peroxidation. Therefore, a strict control of the levels of peroxides is essential for the normal brain growth, maturation and activity. Tayarani and coll. (1989) revealed that there is a large glutathione peroxidase (GPx) activity in adult rat choroid plexuses (CPs). We hypothesize that choroidal GPx activity is essential to control levels of peroxides, like H2O2, in the brain fluid environment during development. We demonstrated by Western Blot and immunohistochemistry that GPx1, GPx4, and glutathione reductase (GR) were consistently expressed in CP epithelial cells of 2 to 17-day-old rats. During that period, GPx and GR specific activities in CP tissue were high compared to the activities measured in cerebral cortex and liver. Rat isolated live CPs were highly efficient at detoxifying H2O2 from mock cerebrospinal fluid (CSF). Mathematical modeling of these results suggested that in vivo, CPs could detoxify a burst of 50 µM of H2O2 in the lateral ventricles within 1 min. The respective contribution of GPxs and catalase in this detoxification process was investigated using a cellular model of the blood-CSF barrier and enzyme inhibitors. We showed that GPxs rather than catalase are responsible for detoxifying concentrations of H2O2 up to 250 µM and that their efficacy is such that H2O2 detoxification is mainly driven by its diffusion rate. These data demonstrate that choroidal GPxs play a pivotal role in the early neuroprotective functions of CPs by controlling peroxides in the brain fluid environment.

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## Effet de corps cétoniques sur la barrière hémato-encéphalique dans le cadre de la Maladie d'Alzheimer

**Pietra Candela,** *Univ. Artois, EA 2465, Laboratoire de la Barrière Hémato-Encéphalique (LBHE), F-62300 Lens, France*

Parmi les stratégies préventives de lutte contre la Maladie d'Alzheimer (MA), la mise en place des protocoles nutritionnels adaptés, suscitent un intérêt croissant. C'est le cas du régime cétogène, déjà utilisé comme un traitement non médicamenteux contre l'épilepsie. Il s'agit d'un régime pauvre en glucides et riche en graisses qui produit dans l'organisme la formation de corps cétoniques (KBs). Il a été démontré que ces dernières ont un effet bénéfique sur la MA par leur effet neuroprotecteur mais également en diminuant le dépôt d'Aß au niveau cérébral. A l'heure actuelle, la plupart des études se concentrent sur la composante neuronale et oublient la barrière hémato-encéphalique (BHE) qui intervient pourtant dans la MA. En utilisant une approche méthodologique mixte, in vivo (microvaisseaux cérébraux extraits de souris 129Sv et in vitro (modèle in vitro de BHE humaine), nous évaluons l'impact de ce régime cétogène et plus particulièrement de KBs sur l'expression des transporteurs et des récepteurs impliqués dans les KBs (MCT1), glucose (GLUT1) et dans le transport et la synthèse des peptides Aβ (BCRP, P-gp, LRP1, RAGE, BACE, PSEN1, PSEN2). Nos résultats montrent que les corps cétoniques modulent la physiologie de la BHE en régulant l'expression de certains transporteurs et récepteurs du peptide Aβ au niveau des cellules endothéliales ainsi que des enzymes synthétisant l'Aβ. La perméabilité de la BHE n'a pas été affectée.

COMMUNICATIONS AFFICHEES

## Prediction of human clinical data based on human primary innovative in vitro models. Part I: The BBB Modelling

**Fabienne Glacial,** *BrainPlotting, IPEPS-ICM, 47 Boulevard de l'Hopital, 75013 Paris*

At BrainPlotting, our main objective is to make Human predictions of brain pharmacokinetics (PK). To that end we have developed a proprietary method: the Brain Exposure Prediction (BEP), based on primary human in vitro tools from fresh human brain resections. With this method, we simulate the unbound drug concentration versus time profiles in brain and plasma following dosing, using in vitro generated PK parameters and physiologically based PK modelling.

In this presentation we focus on the characterization of our BBB models and the expression of tight junction proteins and specific transporters. In partnership with Servier Laboratories, we conducted functional studies on one of our validated BBB model, with influx and efflux permeability measurement of 20 reference molecules. Our BBB models are also used by several academic teams specialized on the BBB.

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## Integrity of blood-brain interfaces in an animal model of severe neonatal jaundice

**Rainui Guy,** *Brain Interfaces Platform, CRNL, INSERM U1028-CNRS UMR5292, Lyon, FR*

Severe jaundice is a disease affecting newborns and characterized by an elevated concentration of bilirubin in the blood. Bilirubin is the breakdown product of hemoglobin released by dying red blood cells. It is converted by the liver glucuronyltransferase enzyme (UGT1A1) into a glucuronylconjugate which is removed from the organism. At birth, this enzyme is immature and plasma protein binding capacity is limited. When bilirubin concentration exceeds albumin binding capacity, free bilirubin enters the central nervous system and cause neurological disorders. The aim of this study is to analyse the effect of postnatal bilirubin increase on the blood-brain interfaces (BBI) integrity, using the hyperbilirubinemic Gunn rat, which is deficient in UGT1A1. We evaluated the blood-brain and blood-CSF permeability to sucrose, a small hydrophilic marker, in two groups of rats, first Gunn rats "jj" and the second one "NN/Nj" healthy rats. Rats received an intraperitoneal injection of [14C]sucrose at birth (P0) or on day 9 (P9). [14C]Sucrose levels in the plasma, the cerebrospinal fluid (CSF) and the brain tissue were measured and permeability constants for the Blood-CSF-Barrier (Kin CSF

= CCSF t / AUC 0->t) and Blood-Brain-Barrier (Kin brain = Cbrain t / AUC 0->t) were calculated. Results showed that neither an acute (P0), nor a sustained (P9) exposure to a high level of free bilirubin damages the permeability of the BBI. On the contrary, we noticed a protective effect at birth on the BBB. Bilirubin induced neurological disorders are not due to an alteration of BBI tight junctions.

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## Connexin 30 is expressed in a subtype of mouse brain pericytes

Noémie Mazaré, *CIRB, Collège de France, CNRS UMR 7241/INSERM U1050*

Pericytes are mural cells of blood microvessels, which play a crucial role at the neurovascular interface of the central nervous system. They are involved in the regulation of blood–brain barrier integrity, angiogenesis, clearance of toxic metabolites, capillary hemodynamic responses, neuroinflammation and they demonstrate stem cell activity. Morphological and molecular studies to characterize brain pericytes recently pointed out some heterogeneity in pericyte population. Nevertheless, a clear definition of pericyte subtypes is still lacking. Here, we demonstrate that a fraction of brain pericytes express Connexin 30 (Cx30), a gap junction protein which, in the brain parenchyma, was thought to be exclusively found in astrocytes. Cx30 could thus be a candidate protein in the composition of the gap junction channel already described between endothelial cells and pericytes, or could act intracellularly to regulate pericyte morphology, as already observed in astrocytes. Altogether, our results suggest that Cx30 defines a novel brain pericyte subtype.

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## Lithium transport at the blood-brain barrier in in vivo rat and in vitro human models

Huilong Luo, *Inserm UMRS1144*

This study deciphers Li+ transport at the BBB focusing on Na+-coupled transporters (NHE (SLC9), NBC (SLC4) and NKCC (SLC12)). The BBB permeability of Li+ evaluated in the rat was 2% that of high passive diffusion compounds. Gene expression of Na+-coupled transporters in hCMEC/D3 cells, human hematopoetic stem cells-derived BBB models (HBLEC) and human primary brain microvascular endothelial cells (hPBMEC) showed the following rank order with close expression profile: NHE1 > NKCC1 > NHE5 > NBCn1 while NHE2, NHE3, NHE4, NBCn2, NBCe1 and NBCe2 mRNA were barely detected. Na+ depletion increased Li+ uptake by 3.3-fold in hCMEC/D3 and Li+ permeability through HBLECs monolayers by 1.6-fold. DMA (NHE inhibitor), DIDS (anionic carriers inhibitor) and bumetanide (NKCC inhibitor) decreased significantly the uptake of Li+ by hCMEC/D3 by 52%, 51% and 47%, respectively while S0859 (NBC inhibitor) increased significantly the uptake of Li+ by 2.3-fold. Zoniporide (NHE1 inhibitor) and siRNA interference against NHE1 had no effect on Li+ uptake by hCMEC/D3 cells. Li+ permeability through HBLEC was significantly increased by DIDS, bumetanide, S0859 and zoniporide. Our study suggests that NHE1 and/or NHE5, NBCn1, and NKCC1 may play a significant role in the Li+ transport across the human BBB and be putative variability factors of Li+ response.

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## Impact of three months ketogenic diet on brain microvessels of TgCRND8 mice

Romain Versele, *Univ. Artois, EA 2465, Laboratoire de la Barrière Hémato-Encéphalique (LBHE), F-62300 Lens,France*

Alzheimer's disease (AD) is a neurodegenerative disease characterized by an abnormal accumulation of β-amyloid (Aβ) peptides at the blood-brain barrier (BBB) level . To date, there are no effective drug therapies for AD and the development of new alternative therapeutic approaches such as the ketogenic diet (KD) should be considered. The KD is based on the production of ketone bodies (KBs) in the blood. Indeed, many studies suggest that KBs have a beneficial effect on AD which could decrease the Aβ brain burden. However, the mechanism of how KBs impact brain microvessels and consequently cerebral Aβ accumulation is still unclear. To get a better understanding, it seems essential to focus on the BBB's receptors, transporters and enzymes that are involved in Aβ peptide transport and metabolism. Wild type 129Sv and transgenic TgCRND8 mice

were maintained on KD or Control Diet for 12 weeks. Body weight was assessed, and microvessel fractions were isolated from the total brain. qPCR analyses were performed to study expression of transporters, receptors and enzymes that are involved in the transport and synthesis of KBs, glucose and Aβ peptide at the BBB level. Our results show an increase in weight of AD mice after a KD of three months and a modulation in the expression of some Aβ transporters and enzymes at the brain microvessels. In particular, in response to KD, the expression levels of these genes return to similar values as the ones of WT mice. This work suggests that it may be possible to modulate Aβ transport and synthesis at the BBB level by controlling dietary intakes.

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