European Journal of Pharmaceutical Sciences 187 (2023) 106462

Contents lists available at ScienceDirect

European Journal of Pharmaceutical Sciences

journal homepage: www.elsevier.com/locate/ejps

Blood-brain barrier perturbations by uremic toxins: Key contributors in chronic kidney disease-induced neurological disorders?

Quentin Faucher [a,*], Thomas K van der Made [a], Elizabeth De Lange [b], Rosalinde Masereeuw [a]

[a] Division of Pharmacology, Utrecht Institute for Pharmaceutical Sciences, Utrecht University, Universiteitsweg 99, 3584 CG Utrecht, the Netherlands
[b] Predictive Pharmacology group, Division of Systems Pharmacology and Pharmacy, Leiden Academic Centre for Drug Research, the Netherlands

ARTICLE INFO

Keywords:
Chronic kidney disease
Kidney-brain crosstalk
Uremic toxins
Blood-brain barrier
Pharmacokinetics

ABSTRACT

Chronic kidney disease is multifactorial and estimated to affect more than 840 million people worldwide constituting a major global health crisis. The number of patients will continue to rise mostly because of the aging population and the increased prevalence of comorbidities such as diabetes and hypertension. Patients with advanced stages display a loss of kidney function leading to an accumulation of, *a.o.* protein-bound uremic toxins that are poorly eliminated by renal replacement therapies. This systemic retention of toxic metabolites, known as the uremic syndrome, affects other organs. Indeed, neurological complications such as cognitive impairment, uremic encephalopathy, and anxiety have been reported in chronic kidney disease patients. Several factors are involved, including hemodynamic disorders and blood-brain barrier (BBB) impairment. The BBB guarantees the exchange of solutes between the blood and the brain through a complex cellular organization and a diverse range of transport proteins. We hypothesize that the increased exposure of the brain to protein-bound uremic toxins is involved in BBB disruption and induces a perturbation in the activity of endothelial membrane transporters. This phenomenon could play a part in the evolution of neurological disorders driven by this kidney-brain crosstalk impairment. In this review, we present chronic kidney disease-induced neurological complications by focusing on the pathological relationship between the BBB and protein-bound uremic toxins. The importance of mechanistically delineating the impact of protein-bound uremic toxins on BBB integrity and membrane drug transporter expression and function in brain endothelial capillary cells is highlighted. Additionally, we put forward current knowledge gaps in the literature.

1. Introduction

Chronic kidney disease (CKD) is a common and long-term disease characterized by a gradual loss of kidney structure and function. The increased prevalence of CKD is likely due to the aging population, Western lifestyle evolution and the significant increase in related diseases such as diabetes and hypertension. Currently, CKD is underdiagnosed mainly because of a lack of symptoms until the advanced stages (Webster et al., 2017). Several advanced CKD patients suffer from the uremic syndrome characterized by a systemic retention of endogenous metabolites, also called uremic toxins, and leading to associated pathologies including neurological disorders, such as cognitive impairment and anxiety (Hamed, 2019). The potential clinical impact of uremic toxins on cerebrovascular and neurological complications associated with CKD has been highlighted (Assem et al., 2018; Liabeuf et al., 2021). The retention of uremic toxins is not only due to a glomerular filtration rate reduction but may additionally be due to the decline in proximal tubule function. Recently, particular attention has been paid to tubular secretion as an underestimated kidney function to be accounted for in CKD progression (Lowenstein and Grantham, 2017). Proximal tubular cells (PTC) express numerous membrane transporters involved in uremic toxins' active elimination from blood into urine, a mechanism particularly effective in the elimination of highly plasma protein-bound uremic toxins (PBUTs), which are poorly eliminated by filtration or dialysis (in kidney failure) (Masereeuw and Verhaar, 2020).

At the cellular level, the increased exposure of uremic toxins to the brain results in brain metabolism dysfunction, disturbance of brain neurotransmitter amino acids balance, vascular autoregulation disturbances and blood-brain barrier (BBB) injury (Hamed, 2019). The BBB plays a central role in CKD brain complications since it is one of the major sites of blood-CNS (central nervous system) exchange. The BBB exhibits a highly complex structure in which cerebral capillary

* Corresponding author at: Division of Pharmacology, Department of Pharmaceutical Sciences, Utrecht University, 3584 CG Utrecht, The Netherlands.
E-mail address: q.g.faucher@uu.nl (Q. Faucher).

https://doi.org/10.1016/j.ejps.2023.106462
Received 19 January 2023; Received in revised form 29 April 2023; Accepted 8 May 2023
Available online 9 May 2023

endothelial cells (BBBec) are tightly connected between themselves, by tight junctions, but also with associated cells, especially pericytes, microglia, and the end-feet of astrocytic glial cells (Abbott et al., 2010). The BBB regulates the exchange of a broad range of molecules (*e.g.* nutrients, ions, toxins and drugs) between the brain and the systemic circulation, coordinated by membrane transporters (Abbott et al., 2010; Morris et al., 2017). Although the involvement of proximal tubular transporters in the urinary excretion of PBUTs is well characterized, much less is known about the effects of PBUTs on the BBB.

We hypothesize that the reduction in proximal tubular transporters' functionality in CKD leads to (1) the systemic retention of PBUTs, (2) a disruption of the BBB integrity and (3) an alteration of the activity of BBBec membrane transporters. This phenomenon could play a part in the evolution of CKD-associated neurological disorders driven by the kidney-brain crosstalk impairment. Moreover, functional impairment of BBBec transporters as well as PBUTs pass-through BBB could be considered as variables for brain physiologically based pharmacokinetic modeling (PBPK) in CKD patients, to provide prediction of drug transport in CKD-induced BBB perturbed conditions. We conducted a non-systematic literature search to compose a narrative review on the research niche of the PBUTs/BBB pathological relationship in the context of CKD-associated neurological disorders. We present (i) the PBUTs blood accumulation phenomena in advanced CKD patients, (ii) an overview of the CKD-associated neurological complications and the involvement of the kidney-brain crosstalk impairment, (iii) the current knowledge on the PBUTs/BBB pathological relationship, (iv) we elaborate on the above-cited hypothesis and finalize (v) discussing the current challenges and needs.

2. Chronic kidney disease-induced neurological disorders

2.1. Advanced chronic kidney disease and blood accumulation of protein-bound uremic toxins

The kidneys act as a biological filtration system of the organism, ensuring the removal of metabolic waste, drugs and toxins from the systemic circulation. This blood detoxification process involves two consecutive structures of the nephron, (i) the glomeruli consisting of biological nanofilters with an approximately 66-kDa molecular weight cut-off and (ii) tubules, ensuring the active secretion of specific products (Fig. S1) (FASN et al., 2015). Patients suffering from CKD present a progressive loss of kidney function, and may only be diagnosed at an advanced stage of the pathology since symptoms are often non-specific at early stages (Webster et al., 2017). The diagnosis is based on estimated glomerular filtration rate (eGFR; <60 mL/min per 1.73 m^2) or kidney damage markers (*e.g.,* albuminuria and abnormalities on histology) evaluation. CKD patients are categorised according to their kidney function impairment and risk of CKD progression. Patients affiliated with the category G5 (eGFR <15 mL/min per 1.73m^2) reached end-stage kidney disease (ESKD) meaning that the kidney is no longer able to sustain its vital function which negatively affects their quality of life. Therefore, these patients resort to kidney replacement therapy including dialysis or kidney transplantation (Webster et al., 2017). Dialysis is the mainstay and most widely used therapy (Wong et al., 2018). Although these renal replacement methods can partly mimic filtration by the glomeruli, they cannot substitute kidney tubules' function. This is particularly true for a heterogeneous group of uremic toxins, the PBUTs (Table S1), which is poorly eliminated by conventional dialysis methods and therefore accumulate into the blood circulation (Masereeuw and Verhaar, 2020). The classification and list of uremic toxins are available via the network EUTox (https://www.uremic-toxins.org/). Drug transporters in the PTC are responsible for the active elimination of the PBUTs in urine. These transporters are expressed at the basolateral side (influx transporters) and the apical side (efflux transporter) of the PTC and ensure the highly-specific and vectorial transepithelial PBUTs excretion. A list of the most studied drug tubular transporters and examples of their PBUT substrates are provided in the Table 1. However, several studies have shown a decrease in the expression and function of tubular transporters in CKD (Torres et al., 2021).

Overall, PBUTs accumulate in the plasma of advanced CKD patients due to kidney impairment and functional alteration of membrane transporters, as well as, limited dialysis capacity. This systemic retention is part of the uremic syndrome, also characterized by multiple organs dysfunction and dysbiosis of the gut microbiota (Nigam and Bush, 2019). It is noteworthy that well-known PBUTs (*e.g.*, indoxyl sulfate (IS), p-cresyl sulfate (pCS), and indole-3-acetic acid (IAA)) are produced by the gut microbiota, and potential changes in intestinal motility, microbiota diversity and molecular environment in CKD could increase the production of uremic toxins precursor (Glorieux et al., 2020).

2.2. Neurological complications associated with CKD-uremic syndrome

The complex systemic and multi-scale metabolic disorder associated with the uremic syndrome promotes a chronic proinflammatory state resulting in multi-organ dysfunction and enhanced cardiovascular risk (Tuegel and Bansal, 2017). Moreover, CKD patients commonly experience neurological complications while patients suffering from ESKD exhibit more than one (Hamed, 2019), indicating a high prevalence of CKD-associated neurological complications, which leads to severe deterioration of quality of life and higher morbidity/mortality. Neurological complications include cognitive impairment, uremic encephalopathy, seizures, anxiety, cerebrovascular stroke, and extrapyramidal movement disorders (*e.g.,* Parkinsonism) (Hamed, 2019). Several factors are involved such as metabolic and hemodynamic disorders, accumulation of uremic toxins and BBB impairment (Jabbari and Vaziri, 2018). Unfortunately, this pathogenesis is still poorly understood. For instance, uremic encephalopathy diagnosis is often based on retrospective evaluation, and relevant defining clinical and laboratory parameters are not used in routine checks yet (Rosner et al., 2022). In a normal physiological state, the kidney-brain crosstalk relies on the neuro-endocrine/kidney interaction (*e.g.,* blood osmolality regulation by vasopressin) and the renal sensory nerve activity (*e.g.,* maintaining sodium balance). Although many kidney-damage factors are likely to contribute to cerebral dysfunction (*e.g.,* impaired renorenal reflex, electrolyte imbalances, oxidative stress and proinflammatory molecules) further studies are still needed to decipher the underlying mechanisms (Tanaka and Okusa, 2020). The uremic toxins appear to be major contributors (Assem et al., 2018; Liabeuf et al., 2021). A clinical study with 199 CKD patients and 84 patients with normal renal function showed a correlation between cognitive function impairment and elevated plasma IS and pCS concentration (Yeh et al., 2016). Some pre-clinical evidence showed PBUTs accumulation in brain tissue. Recently, elevated concentrations of IS and pCS were found in the brain of mice with renal failure compared to control mice (E. Sato et al., 2017). Analysis conducted on postmortem brain tissues, collected from rats exposed to IS for 4 weeks, revealed an accumulation of the toxin in the brainstem, cerebellum, striatum and hippocampus. Moreover, chronic exposure to IS led to animal behavioural alterations (Karbowska et al., 2020). Another study reported a significant increase of pCS concentration in the hippocampus and IS in the frontal lobe, hippocampus and basal ganglion in mice brains after 5/6 nephrectomy with CKD-like pathology. The administration of an oral active carbon absorbant that prevents IS and pCS absorption into the circulation (AST-120) was shown to reduce (i) IS accumulation in the frontal lobe and hippocampus, (ii) hippocampal inflammation, and (iii) cognitive impairment in CKD-like mice (Li et al., 2021). Altogether, these studies show that uremic toxins can reach and accumulate within neural tissue and might influence cognitive functions. However, further research studying their intra-brain distribution and concentration with a large screening panel should be considered, since most literature is available on the two most widely studied toxins, IS and pCS, only. Futhermore, it is worth mentioning that the prevalence of neurological complications is still

Table 1
Main PTC transporters and examples of their pharmaceutical and PBUT substrates.

Transporter Solute carrier (SLC)	Location	Examples of drugs substrates	Examples of PBUT substrates	References
OAT1	Basolateral	Adefovir, tenofovir, hydrochlorothiazide, furosemide, cefazoline	Indoxyl sulfate, kynurenine, hippuric acid	(Ivanyuk et al., 2017; Wu et al., 2017)
OAT3	Basolateral	Bumetanide, furosemide, olmesartan, valsartan, cefazoline	Indoxyl sulfate, p-cresylsulfate, CMPF	(Ivanyuk et al., 2017; Wu et al., 2017)
OCT2	Basolateral	Amyloride, pindolol, procainamide, pramipexole	Putrescine	(Ivanyuk et al., 2017; Masereeuw et al., 2014)
ATP-Binding Cassette (ABC)				
MRP2	Apical	Indomethacin, furosemide, pravastatin	Kynurenic acid, indoxyl sulfate and hippuric acid	(Ivanyuk et al., 2017; Masereeuw et al., 2014)
MRP4	Apical	Cefazoline, hydrochlorothiazide, furosemide, simvastatin	Kynurenic acid, indoxyl sulfate and hippuric acid	(Ivanyuk et al., 2017; Masereeuw et al., 2014)
BCRP	Apical	Sulfasalazine, furosemide, simvastatin	Kynurenic acid, indoxyl sulfate, hippuric acid, p-cresyl sulfate	(Ivanyuk et al., 2017; Masereeuw et al., 2014)

BCRP, breast cancer resistance protein; CMPF, 3-carboxy-4-methyl-5-propyl-2-furanpropionate; MRP, multidrug resistance-associated protein; OAT, organic anion transporter; OCT, organic cation transporter.

high for CKD patients undergoing dialysis. Moreover, dialysis-related factors such as osmolar shifts and pro-inflammatory state further add to the CKD-specific factors suggesting that repeated dialysis may not be a long-term solution (Gupta, 2020; Karunaratne et al., 2018).

A summary of pre-clinical and clinical studies showing direct associations between uremic toxins and neurological complications is available in the above-mentioned review (Liabeuf et al., 2021). Moreover, an in-depth review summarized current knowledge about uremic toxins' involvement in cerebrovascular diseases in CKD patients (Assem et al., 2018). Overall, uremic toxins could have indirect effects on the brain through vascular dysfunction (Six et al., 2015) and hemostasis (Kamiński et al., 2017) as well as direct effects, *i.e.,* neurotoxicity on neuronal cells (Watanabe et al., 2021), neuroinflammation and oxidative stress on glial cells (Adesso et al., 2017; Li et al., 2021), and BBB disruption (Bobot et al., 2020). The BBB is the border crossing between the blood and neural tissue and constitutes the access route for uremic toxins, hence the importance of studying the PBUTs/BBB pathological relationship and putative consequences (Fig. 1). Although we focused mainly on PBUTs, it must be emphasised that other uremic toxins (*e.g.*, creatinine, trimethylamine-N-oxide (TMAO)) can also influence brain functions and hence be considered especially in non-dialysis patients (Hernandez et al., 2022; Liabeuf et al., 2021).

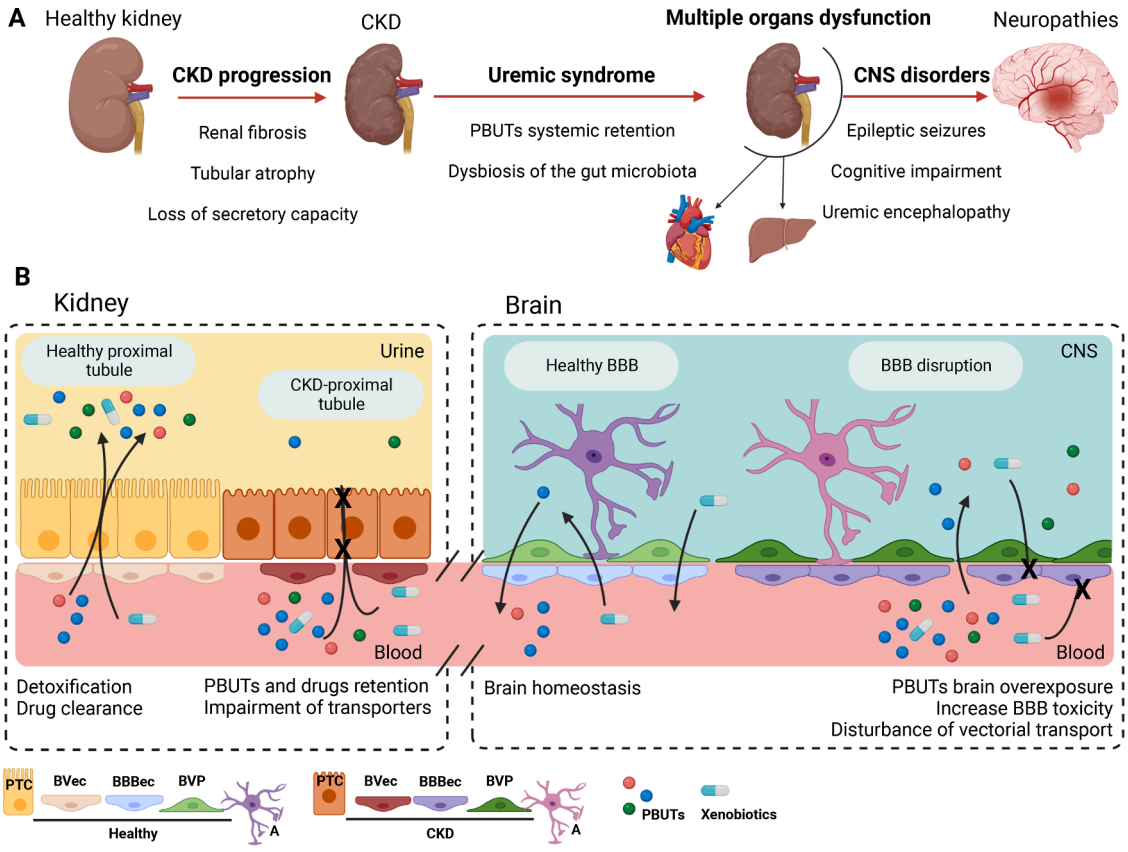

Fig. 1. Kidney-brain crosstalk and putative consequences in CKD-induced neurological complications at the (**A**) anatomical and (**B**) cellular level of tubular and blood-brain barrier. A, astrocytes; BBBec, blood-brain barrier endothelial cell; BVec, blood vessel endothelial cell; BVP, blood vessel pericyte; CKD, chronic kidney disease; CNS, central nervous system; PBUTs, protein-bound uremic toxins; PTC, proximal tubular cell. Illustration created with BioRender.com.

3. Protein-bound uremic toxins/blood-brain-barrier pathological relationship

3.1. Blood-brain barrier structure and function

The BBB refers to the blood vessels that vascularize the CNS and regulate the movements of molecules, playing a crucial role in neuronal function and protection (Profaci et al., 2020) (Fig. 1). The BBB is created by the interaction between the tightly bound BBBec and associated non-neuronal specific cells (*i.e.* end-feet of astrocytic glial cells and pericytes). These cells are part of the neurovascular unit (NVU), formed by neurons, interneurons, astrocytes, basal lamina covered with smooth muscular cells and pericytes, endothelial cells and extracellular matrix (Profaci et al., 2020). The intimate interaction between the several cell types of the NVU results in a functional coupling of neuronal and vascular components. Due to its special properties, the BBB regulates the exchange of a broad range of molecules (*e.g.* nutrients, ions, toxins and drugs) between the brain and the systemic circulation, also coordinated by membrane transporters (Abbott et al., 2010; Banks, 2016; Kadry et al., 2020; Morris et al., 2017).

The BBBec are phenotypically different from other endothelial cells of an organism, by exhibiting continuous non-fenestration due to inter-endothelial cell tight junctions, which decrease paracellular diffusion. Moreover, BBBec, under normal condition, presents a low level of transcytosis implying little transcellular transport capacity. In contrast, the BBBec express many membrane transporters at both the luminal (blood) and abluminal (CNS) side (Table 2). These include different solute carrier-mediated transporters, such as GLUT1 that is involved in glucose exchange, LAT1–2 and EAAT1–3 that govern the transport of specific amino acids such as asparagine, methionine and glutamate, and the nucleoside transporters ENT1–2 for transport of nucleosides. Transporters are also expressed for both anionic (OAT2–3, OATP1A4, OATP2B1) and cationic (OCTN2, OCT1–3) organic ions (Pandit et al., 2020). In addition, the BBBec express several efflux transporters belonging to ATP-Binding Cassette superfamily. Among them, the transporter *ABCB1* (P-glycoprotein (P-gp) or multidrug resistance gene 1 (MDR1)) which is involved in the clearance of many substrates including amyloid-β and drugs (*e.g.,* dexamethasone, verapamil, and vinblastine), breast cancer resistance protein (BCRP; *ABCG2*) responsible of the efflux of sulfoconjugated organic anions and some multidrug resistance-associated proteins (MRPs; *ABCC1,2,4,5*) ensuring the elimination of glucuronides and glutathione conjugates (Mahringer and Fricker, 2016; Morris et al., 2017). These BBBec transporters play a central role in blood-CNS exchange by facilitating the transport of highly-specific molecules, including endogenous metabolites and drugs (Table 2) .

It is worth mentioning that the BBB is a complex structure maintained by a specific cellular/matrix organization. The close interaction between BBBec, pericytes and astrocytes ensures the proper structure, exchange capacities and regulation of the neurovascular microenvironment. Thus, BBB disruption in neurological disease is not an "on-off" mechanism and further research is needed to capture its complexity (Profaci et al., 2020).

Table 2
Main BBBec transporters and their endo/exogenous substrates.

Transporter	Location	Examples of drugs substrates	Putative PBUTs substrates	References
Solute carrier (*SLC*)				
OAT3	Abluminal and luminal	Levofloxacin	Indoxyl sulfate, hippuric acid, indoleacetate, CMPF	(Cen et al., 2023; Deguchi et al., 2006; Ohtsuki et al., 2002)
LAT1	Abluminal and luminal	Gabapentin, pregabalin, L-DOPA, and methyldopa	Kynurenine	(Morris et al., 2017; Walker et al., 2019)
OCT2	Luminal	Fluoxetine	Putrescine	(Masereeuw et al., 2014; M. Wang et al., 2022)
ATP-Binding Cassette (*ABC*)				
MRP2	Luminal	Tacrolimus, sequinavir, vincristine	Kynurenic acid, indoxyl sulfate and hippuric acid	(Mahringer and Fricker, 2016; Masereeuw et al., 2014)
MRP4	Abluminal and luminal	6-mercaptopurine, methotrexate	Kynurenic acid, indoxyl sulfate and hippuric acid	(Mahringer and Fricker, 2016; Masereeuw et al., 2014)
BCRP	Luminal	Lamivudine, anthracyclines, topotecan	Kynurenic acid, indoxyl sulfate, hippuric acid, p-cresyl sulfate	(Mahringer and Fricker, 2016; Masereeuw et al., 2014)

BCRP, breast cancer resistance protein; CMPF, 3-carboxy-4-methyl-5-propyl-2-furanpropionate; LAT, large neutral amino acids transporter; MRP, multidrug resistance-associated protein; OAT, organic anion transporter; OCT, organic cation transporter.

3.2. Impact of uremic toxins on blood-brain-barrier integrity and vectorial transport capability

Uremic toxins can affect the brain microvasculature as well as brain resident cells, including microglia and neurons (Assem et al., 2018). Regarding the BBB, IS was found to induce oxidative stress and apoptosis, and reduced mitochondrial membrane potential in human primary astrocytes (Lin et al., 2019). A release of proinflammatory cytokines (*i.e.,* TNF-α and IL-6) was observed in primary mouse astrocytes exposed to IS (Adesso et al., 2017). Increase production of IL-1β has been reported in human astrocytes treated with quinolinic acid as well as a dose-dependent reduction in glutamine synthetase activity (Ting et al., 2009). Another study showed that human fetal primary astrocytes exhibited an upregulation of chemokine receptor expression following quinolinic acid exposure (Guillemin et al., 2003). A recent review summarized the effect of uremic toxins on endothelial cell integrity highlighting that IS promotes senescence of large-vessel endothelial cells, and generates disruption of contact between pulmonary artery endothelial cells. Moreover, homocysteine was shown to increase H_2O_2 production and triggering apoptosis in endothelial cells (Assem et al., 2018). Another study highlighted that IS and inorganic phosphate reduced NO and induced ROS production in a murine cerebral endothelial cell line (Stinghen et al., 2014). Altogether, these studies suggest a direct toxic effect of uremic toxins on BBBec, however, further studies are needed to clarify this concern.

A few studies have investigated the expression level of BBBec membrane transporters during kidney injury (Fig. 2 and Table S2). A research group identified a significant decrease of Bcrp, Mrp2/4, Oat3, Oatp2/3 and P-gp, at both RNA and protein level, in 5/6 nephrectomized rat brain. Similar findings were observed in rat brain endothelial cells exposed to serum from a rat chronic kidney failure model (Naud et al., 2012). In contrast, a recent study, using isolated brain capillaries from mice ischemia/reperfusion injury models, showed an increase in the level of Bcrp, P-gp and Glut1 protein expressions as well as an increase in the mRNA expression of Pit2, Nhe1 and Cat1 (Burek et al., 2020). This contradiction might be related to the acute (Burek et al., 2020) *versus* chronic (Naud et al., 2012) nature of the kidney injury. However, these results suggest an alteration in the expression of BBBec transporters during kidney failure.

Variations in the expression and functionality of drug transporters and metabolism enzymes have been reported in CKD patients (Fig. 2)

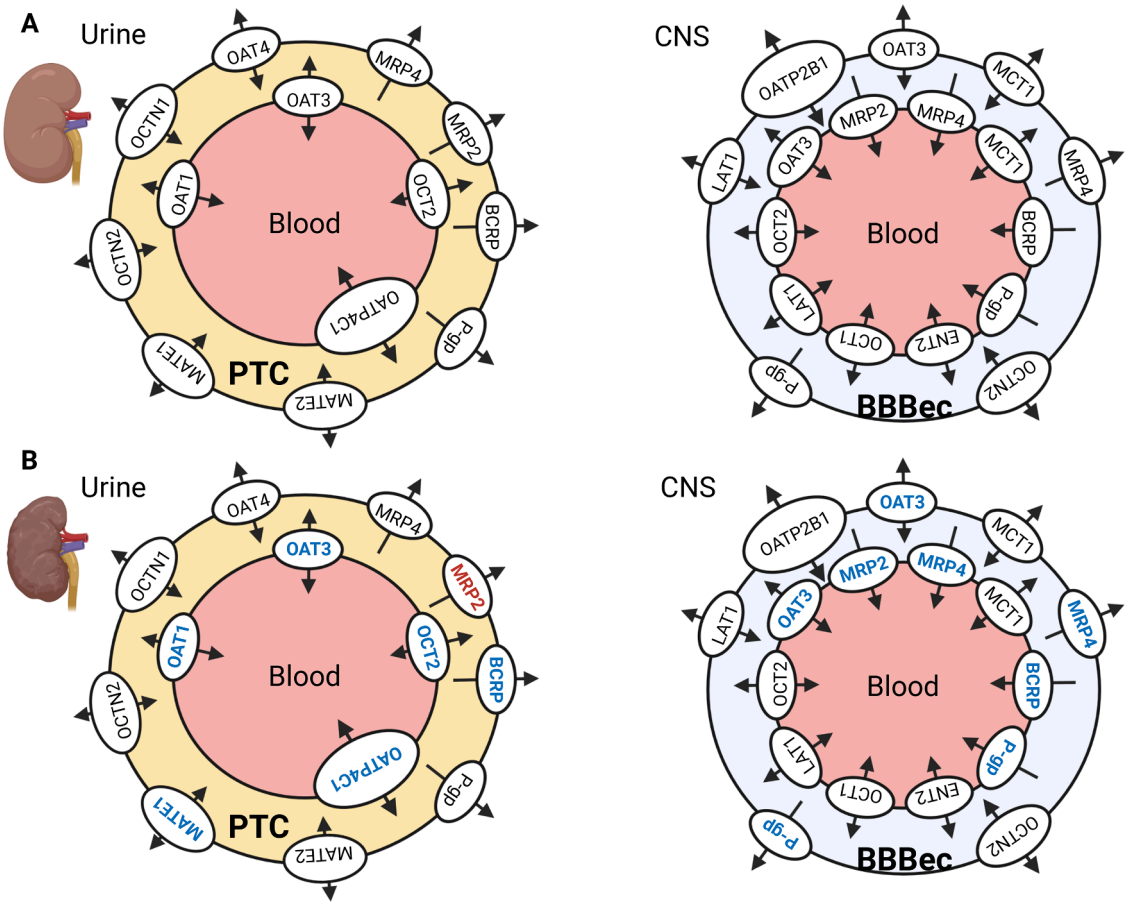

Fig. 2. PTC and BBBec membrane transporters expression profile in (**A**) healthy and (**B**) CKD (red indicates an increase and blue a decrease). BBBec, blood-brain barrier endothelial cell; BCRP, breast cancer resistance protein; ENT, nucleoside transporters; LAT, large neutral amino acids transporter; MATE, multidrug and toxin extrusion; MCT, monocarboxylate transporters; MRP, multidrug resistance-associated protein; OAT, organic anion transporter; OATP, organic anion transporting polypeptides; OCT, organic cation transporter; OCTN, organic cation / carnitine transporter; Pgp, P-glycoprotein; PTC, proximal tubular cell. Illustration created with BioRender.com and based on (Morris et al., 2017; Naud et al., 2012; Torres et al., 2021).

(Torres et al., 2021). A mechanism that could be part of the so-called *Remote Sensing and Signaling Theory* (RSST) *i.e.,* a multi-scale and adaptive communication network based on the expression of multi-specific membrane transporters in many tissues that maintain or even restore homeostasis. Disruption of this network may play a central role in the progression of the uremic syndrome (Nigam et al., 2020; Nigam and Bush, 2019). In healthy kidneys, tubular cells can sense elevated plasma levels of uremic toxins and react by increasing transporter expression levels to support toxin excretion, as was demonstrated for IS regulating OAT1 (Jansen et al., 2019). PBUTs could have a similar effect on the BBBec and, therefore, modulate the expression and function of membrane transporters. In support, human brain microvascular endothelial cells treated with IS or IAA for 48 h in a co-culture model of proximal tubule kidney cells exhibited an increased mRNA expression of *ABCC4/5* and *ABCG2* as well as an IS-dependent decrease of *SLC7A5* encoding for MRP4/5, BCRP and LAT1, respectively. These cells exhibited also an increased protein expression of MRP4 and MCT1 after IS exposure (Burek et al., 2020). Although not discussed in this review, it is worth mentioning that transporters expressed in choroid plexus epithelial cells that form the blood-cerebrospinal fluid (CSF) barrier may also play a key role and should be considered (Morris et al., 2017).

Previous studies have shown that several neurological disorders, including Alzheimer's and Parkinson's disease, are associated with changes in the expression and/or functionality of certain efflux drug transporters (Morris et al., 2017). Furthermore, the inhibition of blood-to-brain transport of kynurenine abrogated the development of depression-like behavior in response to inflammation in a murine model (Walker et al., 2019). Another interesting mechanism reported is the OAT-dependent brain-to-blood efflux. The brain-to-blood transport of IS was evaluated as found by using the brain efflux index method in rats. The authors reported an IS efflux that could be inhibited by diverse molecules such as probenecid (a well-known OAT-inhibitor), benzylpenicillin, and other PBUTs *i.e.,* hippuric acid and CMPF (3-carboxy-4-methyl-5-propyl-2-furanpropionate) (Ohtsuki et al., 2002). They also demonstrated the rOat3-dependent transport characteristic of IS with *Xenopus* oocytes model, as well as, found inhibition of this transport by some metabolites of neurotransmitters such as melatonin and imidazolacetic acid, and drugs like acyclovir, cefazolin, baclofen, 6-mercaptopurine, benzoic acid and ketoprofen (Ohtsuki et al., 2002). Another study with *Xenopus* oocytes model revealed that hippuric acid, indoleacetate and CMPF are substrates of rOat3 while CMPF is also a substrate of rOatp2 (Deguchi et al., 2006).

Altogether, these results suggest that the deleterious effect of PBUTs on the brain could be associated with variations in the expression and activity of membrane transporters. However, further studies need to be undertaken to clarify the expression levels of BBBec transporters in CKD patients, determine the PBUTs-dependent effect on transporters function, and identify specific transporter/PBUTs couples. Such studies are still challenging, given that acquisition of human brain tissue samples remains complex, especially for CKD patients and thus limits the study of human brain endothelial cells. On the another hand, the emergence of advanced *in vitro* BBB models that encompass a controlled microfluidic environment and a cell mixture (endothelial cells, pericytes and astrocytes) represent promosing models for transport analysis and barrier

integrity assessment (Guarino et al., 2023).

PBUTs may compete with co-administered drugs commonly used in CKD management (*i.e.,* angiotensin receptor blockers (losartan and valsartan) and furosemide) for the OAT1-dependent transport, which potentially further compromises the residual tubular function (Mihaila et al., 2020). CKD patients are treated with a plethora of drugs; therefore, drug-toxins interactions at the kidney level could exert widespread and unpredictable effects (André et al., 2022). Given the importance of membrane transporters in the movement of low-permeable compounds, an impairment of BBBec transporters in CKD could lead to PBUTs and transporter-specific neurotransmitters brain retention as well as, influence the drugs' brain distribution, which in turn increases the risk for cerebral dysregulation.

4. Blood-brain-barrier impairment by protein-bound uremic toxins as variable for central nervous system physiologically based pharmacokinetic modeling

4.1. Prediction of drug CNS-PK profiles using physiologically based pharmacokinetic modeling

The prediction of drug pharmacokinetic (PK) profiles in the CNS, by means of physiologically based pharmacokinetic modeling (PBPK), is useful to address potential efficacy and safety concerns of drugs in the pre-clinical and clinical phase of drug development (de Lange & Hammarlund Udenaes, 2022). In addition, patient-population specific PBPK models can help to predict drug or compound disposition in patients by incorporating quantitative pathophysiological changes (Hirasawa et al., 2022). The aim is to predict the right drug concentration at the relevant target site in the CNS. Available *in silico* studies and more complex CNS PBPK models have been discussed elsewhere, some of which incorporated drug efflux transporter activity P-gp and BCRP. Although several CNS PBPK models have been developed for small molecules (de Lange & Hammarlund Udenaes, 2022), this review will illustrate a comprehensive CNS PBPK model: Leiden CNS PBPK predictor V3.0 (LeiCNS-PK3.0) (Saleh et al., 2021). The LeiCNS-PK3.0 integrates system (*e.g.,* interindividual variation, species, health and disease, *in vitro*) and drug-specific (physicochemical properties) data to describe the CNS drug delivery in a mechanistic and physiologically relevant manner. LeiCNS-PK3.0 consists of an empirical plasma model and nine different compartments to describe CNS (*e.g.,* brain microvessels, brain extracellular fluid, brain intracellular fluid, lysosomes, cranial and subarachnoidal cerebrospinal fluid compartments and brain cell membranes). Detailed information on the structure of the model can be found in the above-mentioned article (Saleh et al., 2021). This model has been used to evaluate untested scenarios, *e.g.* physiological parameters reported to be changed in various CNS conditions or the impact of uncertainty in the measurement of values included (Hirasawa et al., 2022), and as "what-if-scenarios" (Saleh & de Lange, 2021).

4.2. PBPK incorporating system changes in CKD and modeling CNS-PK profiles of PBUTs

A recent commentary suggested that the confidence in using PBPK models to extrapolate to the CKD population is currently low due to limited experience (Grimstein et al., 2019). It was discussed that the impact of CKD on important physiological parameters (metabolic enzyme and transporter abundance, albumin-facilitated transport, transporter activity due to accumulation of uremic toxins) is underdeveloped (Grimstein et al., 2019). Adaptation of CNS PBPK models to represent CNS drug delivery in the CKD population, and to predict PBUTs accumulation in the brain, has not been studied to the best of the authors' knowledge. This review aimed to summarize current knowledge on the involvement of key drug transporters in BBB in the active drug transport of PBUTs. Together with quantitative information on drug transporter protein abundance and other physiological changes in CKD, this could give mechanistic insights into the intracellular accumulation of PBUTs and even other uremic toxins in the brain in the CKD population. The inclusion of parameters associated with the PBUTs/BBB relationship (*e.g.,* the intra-brain concentration of PBUTs, expression level of membrane transporters, and drug-toxins interaction) will help to better capture the CNS specificity of CKD patients in order to explore unknown scenarios. Moreover, other parameters representative of BBB breakdown, such as serum biomarkers (*e.g.*, brain-derived neurotropic factor (BDNF), neuron-specific enolase (NSE)) and tight junction protein expression level (*e.g.*, ZO-1 and Claudin-5) (Hernandez et al., 2022) could also be included in a computational predictive model. This comprehensive model could be used for drug (i) discovery/repurposing perspectives (Z. Wang et al., 2020), (ii) dose adjustment in CKD patients and (iii) monitoring for adverse drugs effects as well as (iv) predicting neurotransmitter derangement, and disposition of kidney excreted drugs or their active metabolites, and PBUTs into the CNS.

However, current key knowledge gaps limit individual CNS-PK profiles prediction and extrapolation to specific populations. Information on the quantitative protein abundance of important BBBec drug transporters in humans is currently scarce (de Lange & Hammarlund Udenaes, 2022). Such information can, however, be of great importance to relate transporter protein abundance between *in vitro* systems (Dehouck et al., 2022), animals (Ito et al., 2011) and humans and could also aid to extrapolate data of drugs, that are actively transported, to specific patient populations (R. Sato et al., 2021). Furthermore, more quantitative information on variability in CNS physiology parameters and measured drug concentrations in humans would help to more accurately predict individual CNS-PK profiles. Determination of uremic toxins concentration in the CSF could also provide valuable information on brain exclusion capacity (Mair et al., 2019). Hypothetical modeling and parameter sensitivity analyses suggest, for example, that accurate quantitative estimation of paracellular pore size, pH of ECF and ICF is important as it can impact the rate and extent of passive drug transport across the BBB, as well as distribution within the CNS (brain extra- and intracellular fluids, and cranial and subarachnoidal CSF) in a drug properties dependent fashion (Saleh et al., 2021).

5. Outlook

Facing the high prevalence of neurological complications, there is an urgent need to implement an action focused on CKD-associated neuropathies. Given the systemic, multifactorial and temporal nature of these diseases, it seems worthwhile to coordinate diverse targeted approaches to capture their complexity. Here, we have highlighted a potential target that we believe to be particularly relevant, *i.e.* the PBUTs/BBB relationship and the associated pathological and pharmacological implications. These included an overview of the clinical and pre-clinical evidence of PBUTs brain distribution as well as direct effects on BBB cell components. We reviewed the literature knowledge related to BBBec transporters' expression changes in CKD patients, the evidence of a transporter-dependent brain disposition of PBUTs and put forward the potential impact of membrane transporter impairment on PBUTs brain retention. Finally, we mentioned that the PBUTs systemic concentrations and the concentration-dependent impact of these PBUTs on BBB functionality could be considered as variables, to develop a CDK stage-dependent CNS PBPK model that predicts CNS pharmacokinetics of any solute or drug in CKD patients.

However, current challenges need to be considered such as the vascular to the neuronal transition of PBUTs toxicity. An unknown factor added to the recently proposed neurovascular complex concept (Schaeffer and Iadecola, 2021) encompasses the cellular and molecular diversity of the cerebrovascular tree. More clinical and pre-clinical data about the concentration of a large panel of uremic solutes in CKD brain tissue are still needed as well as the membrane transporter expression profiles. Studies aiming to determine the influx and efflux routes of uremic toxins across the BBB are needed to identify the

transporter/toxin couples. Moreover, the direct effect of uremic toxins on transporter expression should be clarified. The BBB/PBUTs pathological relationship is likely to play a major role in the development and/or aggravation of neurological complications. However, the direct (*e.g.,* toxicity, neuroinflammation) and indirect (*e.g.,* vascular dysfunction, membrane transporters function and co-interaction) implications of PBUTs must be deciphered to gain insight into the CKD-associated neurological complication pathogenesis.

CRediT authorship contribution statement

Quentin Faucher: Conceptualization, Writing – original draft, Writing – review & editing, Funding acquisition. **Thomas K van der Made:** Conceptualization, Writing – original draft, Writing – review & editing. **Elizabeth De Lange:** Conceptualization, Validation, Writing – review & editing, Supervision. **Rosalinde Masereeuw:** Conceptualization, Validation, Writing – review & editing, Supervision, Funding acquisition.

Declartion of Competing Interest

The authors declare no competing interests.

Data availability

Data will be made available on request.

Acknowledgements

The authors are grateful to Marta Garcia Valverde for her valuable input on the figures' design and Joshua Elford for correcting English. RM and QF acknowledge the Horizon Europe Framework Programme (HORIZON) under Marie Skłodowska-Curie grant agreement BBB-UT No. 101107439. RM and Th.K.vd.M received co-funding by the PPP Allowance made available by SGF and Health~Holland, Top Sector Life Sciences & Health, the Netherlands (grant no. LSHM20045-HSGF) to stimulate public-private partnerships.

Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.ejps.2023.106462.

References

Abbott, N.J., Patabendige, A.A.K., Dolman, D.E.M., Yusof, S.R., Begley, D.J., 2010. Structure and function of the blood–brain barrier. Neurobiol. Dis. 37 (1), 13-25. https://doi.org/10.1016/j.nbd.2009.07.030.

Adesso, S., Magnus, T., Cuzzocrea, S., Campolo, M., Rissiek, B., Paciello, O., Autore, G., Pinto, A., Marzocco, S., 2017. Indoxyl sulfate affects glial function increasing oxidative stress and neuroinflammation in chronic kidney disease : interaction between astrocytes and microglia. Front. Pharmacol. 8, 370. https://doi.org/10.3389/fphar.2017.00370.

André, C., Choukroun, G., Bennis, Y., Kamel, S., Lemaire-Hurtel, A.S., Masmoudi, K., Bodeau, S., Liabeuf, S., 2022. Potential interactions between uraemic toxins and drugs : an application in kidney transplant recipients treated with calcineurin inhibitors. Nephrol. Dialysis Transplant.: Off. Public. Eur. Dialysis Transplant Assoc. - Eur. Renal Assoc. 37 (11), 2284-2292. https://doi.org/10.1093/ndt/gfab114.

Assem, M., Lando, M., Grissi, M., Kamel, S., Massy, Z.A., Chillon, J.-.M., Hénaut, L., 2018. The impact of uremic toxins on cerebrovascular and cognitive disorders. Toxins (Basel) 10 (7), 303. https://doi.org/10.3390/toxins10070303.

Banks, W.A., 2016. From blood–brain barrier to blood–brain interface : new opportunities for CNS drug delivery. Nat. Rev. Drug Discovery 15 (4), 4. https://doi.org/10.1038/nrd.2015.21.

Bobot, M., Thomas, L., Moyon, A., Fernandez, S., McKay, N., Balasse, L., Garrigue, P., Brige, P., Chopinet, S., Poitevin, S., Cérini, C., Brunet, P., Dignat-George, F., Burtey, S., Guillet, B., Hache, G., 2020. Uremic toxic blood-brain barrier disruption mediated by AhR activation leads to cognitive impairment during experimental renal dysfunction. J. Am. Soc. Nephrol. 31 (7), 1509-1521. https://doi.org/10.1681/ASN.2019070728.

Burek, M., Burmester, S., Salvador, E., Möller-Ehrlich, K., Schneider, R., Roewer, N., Nagai, M., Förster, C.Y., 2020. Kidney ischemia/reperfusion injury induces changes in the drug transporter expression at the blood–brain barrier *in vivo* and *in vitro*. Front. Physiol. 11 https://doi.org/10.3389/fphys.2020.569881.

Cen, Y., Shan, Y., Zhao, J., Xu, X., Nie, Z., Zhang, J., 2023. Multiple drug transporters contribute to the brain transfer of levofloxacin. CNS Neurosci. Ther. 29 (1), 445-457. https://doi.org/10.1111/cns.13989.

Deguchi, T., Isozaki, K., Yousuke, K., Terasaki, T., Otagiri, M., 2006. Involvement of organic anion transporters in the efflux of uremic toxins across the blood–brain barrier. J. Neurochem. 96 (4), 1051-1059. https://doi.org/10.1111/j.1471-4159.2005.03550.x.

Dehouck, M.-P., Tachikawa, M., Hoshi, Y., Omori, K., Maurage, C.-A., Strecker, G., Dehouck, L., Boucau, M.-C., Uchida, Y., Gosselet, F., Terasaki, T., Karamanos, Y., 2022. Quantitative targeted absolute proteomics for better characterization of an in vitro human blood-brain barrier model derived from hematopoietic stem cells. Cells 11 (24), 3963. https://doi.org/10.3390/cells11243963.

de Lange, E.C.M., Hammarlund Udenaes, M., 2022. Understanding the blood-brain barrier and beyond : challenges and opportunities for novel CNS therapeutics. Clin. Pharmacol. Ther. 111 (4), 758-773. https://doi.org/10.1002/cpt.2545.

FASN, K.S.M.F., MD, G.M.C., MD, P.A.M., FRCP, M.W.T.Mbc.Mm.M.F., MD, A.S.L.Y, 2015. Brenner and Rector's The Kidney, 10 edition. Elsevier. (2 Volume Set), 10e.

Glorieux, G., Gryp, T., Perna, A., 2020. Gut-derived metabolites and their role in immune dysfunction in chronic kidney disease. Toxins (Basel) 12 (4). https://doi.org/10.3390/toxins12040245.

Grimstein, M., Yang, Y., Zhang, X., Grillo, J., Huang, S.-.M., Zineh, I., Wang, Y., 2019. Physiologically based pharmacokinetic modeling in regulatory science : an update from the U.S. food and drug administration's office of clinical pharmacology. J. Pharm. Sci. 108 (1), 21-25. https://doi.org/10.1016/j.xphs.2018.10.033.

Guarino, V., Zizzari, A., Bianco, M., Gigli, G., Moroni, L., Arima, V., 2023. Advancements in modelling human blood brain-barrier on a chip. Biofabrication 15 (2), 022003. https://doi.org/10.1088/1758-5090/acb571.

Guillemin, G.J., Croitoru-Lamoury, J., Dormont, D., Armati, P.J., Brew, B.J., 2003. Quinolinic acid upregulates chemokine production and chemokine receptor expression in astrocytes. Glia 41 (4), 371-381. https://doi.org/10.1002/glia.10175.

Gupta, N., 2020. Brain and kidney crosstalk. In: Prabhakar, H., Gupta, N. (Eds.), Brain and Kidney Crosstalk, Éds. Springer, p. 89-117. https://doi.org/10.1007/978-981-15-2325-0_3.

Hamed, S.A., 2019. Neurologic conditions and disorders of uremic syndrome of chronic kidney disease : presentations, causes, and treatment strategies. Expert Rev. Clin. Pharmacol. 12 (1), 61-90. https://doi.org/10.1080/17512433.2019.1555468.

GOING-FWD Collaborators, Hernandez, L., Ward, L.J., Arefin, S., Ebert, T., Laucyte-Cibulskiene, A., Heimbürger, O., Barany, P., Wennberg, L., Stenvinkel, P., Kublickiene, K, 2022. Blood-brain barrier and gut barrier dysfunction in chronic kidney disease with a focus on circulating biomarkers and tight junction proteins. Sci. Rep. 12 (1), 4414. https://doi.org/10.1038/s41598-022-08387-7.

Hirasawa, M., Saleh, M.A.A., de Lange, E.C.M., 2022. The extension of the LeiCNS-PK3.0 model in combination with the "handshake" approach to understand brain tumor pathophysiology. Pharm. Res. 39 (7), 1343-1361. https://doi.org/10.1007/s11095-021-03154-1.

Ito, K., Uchida, Y., Ohtsuki, S., Aizawa, S., Kawakami, H., Katsukura, Y., Kamiie, J., Terasaki, T., 2011. Quantitative membrane protein expression at the blood-brain barrier of adult and younger cynomolgus monkeys. J. Pharm. Sci. 100 (9), 3939-3950. https://doi.org/10.1002/jps.22487.

Ivanyuk, A., Livio, F., Biollaz, J., Buclin, T., 2017. Renal drug transporters and drug interactions. Clin. Pharmacokinet. 56 (8), 825-892. https://doi.org/10.1007/s40262-017-0506-8.

Jabbari, B., Vaziri, N.D., 2018. The nature, consequences, and management of neurological disorders in chronic kidney disease. In: Hemodialysis International. International Symposium on Home Hemodialysis, 22, p. 150-160. https://doi.org/10.1111/hdi.12587.

Jansen, J., Jansen, K., Neven, E., Poesen, R., Othman, A., van Mil, A., Sluijter, J., Sastre Torano, J., Zaal, E.A., Berkers, C.R., Esser, D., Wichers, H.J., van Ede, K., van Duursen, M., Burtey, S., Verhaar, M.C., Meijers, B., Masereeuw, R., 2019. Remote sensing and signaling in kidney proximal tubules stimulates gut microbiome-derived organic anion secretion. Proc. Natl. Acad. Sci. USA 116 (32). https://doi.org/10.1073/pnas.1821809116, 16105-16110.

Kadry, H., Noorani, B., Cucullo, L., 2020. A blood–brain barrier overview on structure, function, impairment, and biomarkers of integrity. Fluids Barriers CNS 17 (1), 69. https://doi.org/10.1186/s12987-020-00230-3.

Kamiński, T.W., Pawlak, K., Karbowska, M., Myśliwiec, M., Pawlak, D., 2017. Indoxyl sulfate—The uremic toxin linking hemostatic system disturbances with the prevalence of cardiovascular disease in patients with chronic kidney disease. BMC Nephrol. 18 (1), 35. https://doi.org/10.1186/s12882-017-0457-1.

Karbowska, M., Hermanowicz, J.M., Tankiewicz-Kwedlo, A., Kalaska, B., Kaminski, T.W., Nosek, K., Wisniewska, R.J., Pawlak, D., 2020. Neurobehavioral effects of uremic toxin-indoxyl sulfate in the rat model. Sci. Rep. 10 (1), 9483. https://doi.org/10.1038/s41598-020-66421-y.

Karunaratne, K., Taube, D., Khalil, N., Perry, R., Malhotra, P.A., 2018. Neurological complications of renal dialysis and transplantation. Pract Neurol 18 (2), 115-125. https://doi.org/10.1136/practneurol-2017-001657.

Li, L.-.C., Chen, W.-.Y., Chen, J.-.B., Lee, W.-.C., Chang, C.-.C., Tzeng, H.-.T., Huang, C.-.C., Chang, Y.-.J., Yang, J.-.L., 2021. The AST-120 recovers uremic toxin-induced cognitive deficit via NLRP3 inflammasome pathway in astrocytes and microglia. Biomedicines 9 (9), 1252. https://doi.org/10.3390/biomedicines9091252.

Liabeuf, S., Pepin, M., Franssen, C.F.M., Viggiano, D., Carriazo, S., Gansevoort, R.T., Gesualdo, L., Hafez, G., Malyszko, J., Mayer, C., Nitsch, D., Ortiz, A., Pešić, V.,

Wiecek, A., Massy, Z.A., 2021. Chronic kidney disease and neurological disorders : are uraemic toxins the missing piece of the puzzle? Nephrol. Dialysis Transplant.: Off. Public. Eur. Dialysis Transplant. Assoc. - Eur. Renal Assoc. https://doi.org/10.1093/ndt/gfab223 gfab223.

Lin, Y.-T., Wu, P.-H., Tsai, Y.-C., Hsu, Y.-L., Wang, H.Y., Kuo, M.-C., Kuo, P.-L., Hwang, S.-J., 2019. Indoxyl sulfate induces apoptosis through oxidative stress and mitogen-activated protein kinase signaling pathway inhibition in human astrocytes. J. Clin. Med. 8 (2), 191. https://doi.org/10.3390/jcm8020191.

Lowenstein, J., Grantham, J.J., 2017. Residual renal function : a paradigm shift. Kidney Int. 91 (3), 561-565. https://doi.org/10.1016/j.kint.2016.09.052.

Mahringer, A., Fricker, G., 2016. ABC transporters at the blood–brain barrier. Expert Opin. Drug Metab. Toxicol. 12 (5), 499-508. https://doi.org/10.1517/17425255.2016.1168804.

Mair, R.D., Nguyen, H., Huang, T.-T., Plummer, N.S., Sirich, T.L., Meyer, T.W., 2019. Accumulation of uremic solutes in the cerebrospinal fluid in experimental acute renal failure. Am. J. Physiol. - Renal Physiol. 317 (2), F296-F302. https://doi.org/10.1152/ajprenal.00100.2019.

Masereeuw, R., Mutsaers, H.A.M., Toyohara, T., Abe, T., Jhawar, S., Sweet, D.H., Lowenstein, J., 2014. The kidney and uremic toxin removal : glomerulus or tubule? Semin. Nephrol. 34 (2), 191-208. https://doi.org/10.1016/j.semnephrol.2014.02.010.

Masereeuw, R., Verhaar, M.C., 2020. Innovations in approaches to remove uraemic toxins. Nat. Rev. Nephrol. https://doi.org/10.1038/s41581-020-0299-0.

Mihaila, S.M., Faria, J., Stefens, M.F.J., Stamatialis, D., Verhaar, M.C., Gerritsen, K.G.F., Masereeuw, R., 2020. Drugs commonly applied to kidney patients may compromise renal tubular uremic toxins excretion. Toxins (Basel) 12 (6). https://doi.org/10.3390/toxins12060391.

Morris, M.E., Rodriguez-Cruz, V., Felmlee, M.A., 2017. SLC and ABC transporters : expression, localization, and species differences at the blood-brain and the blood-cerebrospinal fluid barriers. AAPS J. 19 (5), 1317-1331. https://doi.org/10.1208/s12248-017-0110-8.

Naud, J., Laurin, L.-P., Michaud, J., Beauchemin, S., Leblond, F.A., Pichette, V., 2012. Effects of chronic renal failure on brain drug transporters in rats. Drug Metab. Dispos. 40 (1), 39-46. https://doi.org/10.1124/dmd.111.041145.

Nigam, S.K., Bush, K.T., 2019. Uraemic syndrome of chronic kidney disease : altered remote sensing and signalling. Nat. Rev. Nephrol. 15 (5), 301-316. https://doi.org/10.1038/s41581-019-0111-1.

Nigam, S.K., Bush, K.T., Bhatnagar, V., Poloyac, S.M., Momper, J.D., 2020. The systems biology of drug metabolizing enzymes and transporters : relevance to quantitative systems pharmacology. Clin. Pharmacol. Ther. 108 (1), 40-53. https://doi.org/10.1002/cpt.1818.

Ohtsuki, S., Asaba, H., Takanaga, H., Deguchi, T., Hosoya, K., Otagiri, M., Terasaki, T., 2002. Role of blood-brain barrier organic anion transporter 3 (OAT3) in the efflux of indoxyl sulfate, a uremic toxin : its involvement in neurotransmitter metabolite clearance from the brain. J. Neurochem. 83 (1), 57-66. https://doi.org/10.1046/j.1471-4159.2002.01108.x.

Pandit, R., Chen, L., Götz, J., 2020. The blood-brain barrier : physiology and strategies for drug delivery. Adv. Drug Deliv. Rev. 165-166, 1-14. https://doi.org/10.1016/j.addr.2019.11.009.

Profaci, C.P., Munji, R.N., Pulido, R.S., Daneman, R., 2020. The blood–brain barrier in health and disease : important unanswered questions. J. Exp. Med. 217 (4), e20190062 https://doi.org/10.1084/jem.20190062.

Rosner, M.H., Husain-Syed, F., Reis, T., Ronco, C., Vanholder, R., 2022. Uremic encephalopathy. Kidney Int. 101 (2), 227-241. https://doi.org/10.1016/j.kint.2021.09.025.

Saleh, M.A.A., de Lange, E.C.M., 2021. Impact of CNS diseases on drug delivery to brain extracellular and intracellular target sites in human : a “WHAT-IF” simulation study. Pharmaceutics 13 (1), 1. https://doi.org/10.3390/pharmaceutics13010095.

Saleh, M.A.A., Loo, C.F., Elassaiss-Schaap, J., De Lange, E.C.M., 2021. Lumbar cerebrospinal fluid-to-brain extracellular fluid surrogacy is context-specific : insights from LeiCNS-PK3.0 simulations. J. Pharmacokinet. Pharmacodyn. 48 (5), 725-741. https://doi.org/10.1007/s10928-021-09768-7.

Sato, E., Saigusa, D., Mishima, E., Uchida, T., Miura, D., Morikawa-Ichinose, T., Kisu, K., Sekimoto, A., Saito, R., Oe, Y., Matsumoto, Y., Tomioka, Y., Mori, T., Takahashi, N., Sato, H., Abe, T., Niwa, T., Ito, S., 2017. Impact of the oral adsorbent AST-120 on organ-specific accumulation of uremic toxins : LC-MS/MS and MS imaging techniques. Toxins (Basel) 10 (1), 19. https://doi.org/10.3390/toxins10010019.

Sato, R., Ohmori, K., Umetsu, M., Takao, M., Tano, M., Grant, G., Porter, B., Bet, A., Terasaki, T., Uchida, Y., 2021. An atlas of the quantitative protein expression of anti-epileptic-drug transporters, metabolizing enzymes and tight junctions at the blood–brain barrier in epileptic patients. Pharmaceutics 13 (12), 12. https://doi.org/10.3390/pharmaceutics13122122.

Schaeffer, S., Iadecola, C., 2021. Revisiting the neurovascular unit. Nat. Neurosci. 24 (9), 9. https://doi.org/10.1038/s41593-021-00904-7.

Six, I., Gross, P., Rémond, M.C., Chillon, J.M., Poirot, S., Drueke, T.B., Massy, Z.A., 2015. Deleterious vascular effects of indoxyl sulfate and reversal by oral adsorbent AST-120. Atherosclerosis 243 (1), 248-256. https://doi.org/10.1016/j.atherosclerosis.2015.09.019.

Stinghen, A.E.M., Chillon, J.-M., Massy, Z.A., Boullier, A., 2014. Differential effects of indoxyl sulfate and inorganic phosphate in a murine cerebral endothelial cell line (bEnd.3). Toxins (Basel) 6 (6), 6. https://doi.org/10.3390/toxins6061742.

Tanaka, S., Okusa, M.D., 2020. Crosstalk between the nervous system and the kidney. Kidney Int. 97 (3), 466-476. https://doi.org/10.1016/j.kint.2019.10.032.

Ting, K.K., Brew, B.J., Guillemin, G.J., 2009. Effect of quinolinic acid on human astrocytes morphology and functions : implications in Alzheimer’s disease. J. Neuroinflammation 6, 36. https://doi.org/10.1186/1742-2094-6-36.

Torres, A.M., Dnyanmote, A.V., Granados, J.C., Nigam, S.K., 2021. Renal and non-renal response of ABC and SLC transporters in chronic kidney disease. Expert Opin. Drug Metab. Toxicol. 17 (5), 515-542. https://doi.org/10.1080/17425255.2021.1899159.

Tuegel, C., Bansal, N., 2017. Heart failure in patients with kidney disease. Heart 103 (23), 1848-1853. https://doi.org/10.1136/heartjnl-2016-310794.

Walker, A.K., Wing, E.E., Banks, W.A., Dantzer, R., 2019. Leucine competes with kynurenine for blood-to-brain transport and prevents lipopolysaccharide-induced depression-like behavior in mice. Mol. Psychiatry 24 (10), 1523-1532. https://doi.org/10.1038/s41380-018-0076-7.

Wang, M., Sun, Y., Hu, B., He, Z., Chen, S., Qi, D., An, H., Wei, Y., 2022. Organic cation transporters are involved in fluoxetine transport across the blood-brain barrier *in vivo* and *in vitro*. Curr Drug Deliv 19 (4), 508-517. https://doi.org/10.2174/1567201818666210708122326.

Wang, Z., Vilekar, P., Huang, J., Weaver, D.F., 2020. Furosemide as a probe molecule for the treatment of neuroinflammation in Alzheimer’s disease. ACS Chem. Neurosci. 11 (24), 4152-4168. https://doi.org/10.1021/acschemneuro.0c00445.

Watanabe, K., Sato, E., Mishima, E., Watanabe, M., Abe, T., Takahashi, N., Nakayama, M., 2021. Effect of uremic toxins on hippocampal cell damage : analysis *in vitro* and in rat model of chronic kidney disease. Heliyon 7 (2), e06221. https://doi.org/10.1016/j.heliyon.2021.e06221.

Webster, A.C., Nagler, E.V., Morton, R.L., Masson, P., 2017. Chronic kidney disease. Lancet 389 (10075), 1238-1252. https://doi.org/10.1016/S0140-6736(16)32064-5.

Wong, B., Ravani, P., Oliver, M.J., Holroyd-Leduc, J., Venturato, L., Garg, A.X., Quinn, R. R., 2018. Comparison of patient survival between hemodialysis and peritoneal dialysis among patients eligible for both modalities. Am. J. Kidney Dis.: Off. J. Natl.l Kidney Foundation 71 (3), 344-351. https://doi.org/10.1053/j.ajkd.2017.08.028.

Wu, W., Bush, K.T., Nigam, S.K., 2017. Key role for the organic anion transporters, OAT1 and OAT3, in the *in vivo* handling of uremic toxins and solutes. Sci. Rep. 7 (1), 4939. https://doi.org/10.1038/s41598-017-04949-2.

Yeh, Y.-C., Huang, M.-F., Liang, S.-S., Hwang, S.-J., Tsai, J.-C., Liu, T.-L., Wu, P.-H., Yang, Y.-H., Kuo, K.-C., Kuo, M.-C., Chen, C.-S., 2016. Indoxyl sulfate, not p-cresyl sulfate, is associated with cognitive impairment in early-stage chronic kidney disease. Neurotoxicology 53, 148-152. https://doi.org/10.1016/j.neuro.2016.01.006.