Life Sciences 233 (2019) 116684

Contents lists available at ScienceDirect

Life Sciences

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

Review article

Potential therapeutic implications of ergogenic compounds on pathophysiology induced by traumatic brain injury: A narrative review

Gustavo Cassol[b,e], Douglas Buchmann Godinho[b,e], Viviane Nogueira de Zorzi[e], Juliano Boufleur Farinha[b,e], Iuri Domingues Della-Pace[a], Muryel de Carvalho Gonçalves[d], Mauro Schneider Oliveira[a], Ana Flavia Furian[a], Michele Rechia Fighera[c,e], Luiz Fernando Freire Royes[a,b,e,*]

[a] Centro de Ciências da Saúde, Programa de Pós-Graduação em Farmacologia - Universidade Federal de Santa Maria, 97105-900 Santa Maria, RS, Brazil
[b] Programa de Pós-Graduação em Educação Física, Universidade Federal de Santa Maria, 97105-900 Santa Maria, RS, Brazil
[c] Centro de Ciências da Saúde, Departamento de Clínica Médica e Pediatria - Universidade Federal de Santa Maria, 97105-900 Santa Maria, RS, Brazil
[d] Centro de Ciências Biológicas, Departamento de Farmacologia -Universidade Federal do Paraná, 81531-980 Curitiba, PR, Brazil
[e] Laboratório de Bioquímica do Exercício - Universidade Federal de Santa Maria, 97105-900 Santa Maria, RS, Brazil

ARTICLE INFO

Keywords:
TBI
Ergogenic aid
Energetic metabolism
Neuroplasticity
Mitochondrial function

ABSTRACT

Traumatic brain injury (TBI) is a devastating condition that often triggers a sequel of neurological disorders that can last throughout lifespan. From a metabolic viewpoint, the compromising of the energy metabolism of the brain has produced evidence linking the severity of brain injury to the extent of disturbances in the cerebral metabolism. The cerebral metabolic crisis, however, displays that regional heterogeneity varies temporally post-injury. It is important to note that energy generation and mitochondrial function are closely related and interconnected with delayed secondary manifestations of brain injury, including early neuromotor dysfunction, cognitive impairment, and post-traumatic epilepsy (PTE). Given the extent of post-traumatic changes in neuronal function and the possibility of amplifying secondary cascades, different therapies designed to minimize damage and retain/restore cellular function after TBI are currently being studied. One of the possible strategies may be the inclusion of ergogenic compounds, which is a class of supplements that typically includes ingredients used by athletes to enhance their performance. The combination of these compounds offers specific physiological advantages, which include enhanced energy availability/metabolism and improved buffering capacity. However, the literature on their effects in certain biological systems and neurological diseases, such as TBI, has yet to be determined. Thus, the present review aims to discuss the role of ergogenic compounds popularly used in secondary damage induced by this neurological injury. In this narrative review, we also discuss how the results from animal studies can be applied to TBI clinical settings.

1. Introduction

Traumatic brain injury (TBI) causes significant behavioral/emotional disabilities and long-term medical complications. Around 69 million individuals worldwide are estimated to suffer from TBI each year [1]. It is estimated that 1.7 million TBIs/year occur in the United States of America (USA), in which around 50,000 people die and 275,000 are hospitalized [2]. In the European Union, brain injury accounts for 2.5 million people each year, causing 1 million hospital admissions and 75,000 deaths [3,4]. This condition is characterized by a combination of immediate mechanical dysfunction of brain tissue and secondary damage developed over a longer period of time following the injury [5]. The attention given to mild traumatic brain injury (mTBI), which is also known as a concussion, has increased significantly in recent years due to high incidence in professional contact sports. Epidemiological studies have demonstrated that 3.8 million athletes suffer concussions in sports including boxing, American Football, and Australian Rules Football (AFL), with more than half of the retired professional AFL players reporting multiple concussions throughout their playing career [6,7].

Regarding TBI-induced severity, it is important to note that metabolic and functional deficits after neuronal injury are interrelated

* Corresponding author at: Departamento de Métodos e Técnicas Desportivas, Centro de Educação Física e Desportos, Universidade Federal de Santa Maria, 97105-900 Santa Maria, RS, Brazil.
E-mail address: nandoroyes@yahoo.com.br (L.F.F. Royes).

https://doi.org/10.1016/j.lfs.2019.116684
Received 4 June 2019; Accepted 22 July 2019
Available online 24 July 2019
0024-3205/

events. Furthermore, unexpected neurological consequences characterized by late cognitive decline and/or neuropsychiatric performance, many years after neuronal injury, reinforce the idea of a dynamic element underlying the pathophysiology of TBI [5]. For instance, it is well known that the variations of cerebral ATP and mitochondrial phosphorylation capacity (measured by the ATP/ADP ratio) are influenced by TBI severity. However, patients who survive TBI show early activation of the transcriptional program for mitochondrial biogenesis, whereas failure to activate the program leads to reduced mitochondrial content and cellular energetic failure [13], which suggests that mitochondria can be instrumental both in cell failure and cell recovery after neuronal injury.

In support of this perspective, the inclusion of therapeutic ergogenic compounds, which is a class of supplements with specific physiological advantages, in treating neurological diseases has included enhanced energy availability/metabolism and improved buffering capacity [8]. However, their potential effects on TBI pathophysiology require deeper analysis. Thus, this review focuses on the impact of ergogenic compounds on energetic metabolism and the consequences for brain function after TBI (Table 1).

2. TBI pathophysiology

Clinically, traumatic brain injuries (TBIs) are classified by severity: mild, moderate, and severe. Brain damage in TBI is caused by a combination of a primary mechanical injury that initiates a cycle of secondary injury effects that include both systemic complications and cellular injury mechanisms [9,10]. From a metabolic viewpoint, a crucial event that triggers neural dysfunction is the increased release of excitatory amino acids, particularly glutamate. TBI-induced excitotoxicity involves the excessive release of glutamate from presynaptic nerve terminals and reversal of astrocytic glutamate uptake, when there is excessive neuronal depolarization. *N*-methyl-D-aspartate (NMDA) receptors (glutamate receptor subtype) are activated in postsynaptic neurons, opening their receptor-operated cation channels to allow calcium (Ca^{2+}) influx. Elevated intracellular Ca^{2+} levels trigger many cellular pathways, such as the activation of phospholipases including calcineurin, proteases including calpains and caspases, transcription factors including c-Fos, c-Jun and c-myc, nitric oxide synthase (NOS), as well as DNA degrading endonucleases, which characterize secondary injury mechanisms [11].

Increased extracellular glutamate concentrations also augment cerebral glucose utilization. In order to promote the re-establishment of ionic homeostasis, relative hyperglycemia induces extracellular lactate accumulation [12]. Lactate is the central metabolite in cerebral energy substrate delivery, since it is the glycolysis product and main substrate for mitochondrial respiration [13]. In this sense, the lactate/pyruvate ratio has important diagnostic value to distinguish between ischemia and mitochondrial dysfunction in TBI patients [14]. In fact, deficits in mitochondrial bioenergetics occur rapidly following experimental TBI (< 1 h) and may persist for up to 14 days [15]. Following neuronal injury, excessive intracellular Ca^{2+} can exhaust the calcium buffering ability of mitochondria, causing mitochondrial permeability transport pore (mPTP) formation. Consequences of mPTP may include loss of membrane potential and consequent electron transport uncoupling from adenosine triphosphate (ATP) production, in addition to the release of pro apoptotic factors and cell death pathways [16]. Additionally, mPTP formation leads to the production of reactive oxygen species (ROS) and free radicals, which may severely damage cells.

The term “hyperglycolysis” is used to connote the extent of metabolic crisis following injury, since cerebral glucose is rapidly depleted leading to cerebral lactate production. Moreover, brief periods of hyperglycolysis in the acute phase (i.e., first few days postinjury) are followed by global reduction in glycolysis until recovery (weeks to months postinjury) in humans [17]. Metabolic changes following TBI are measured by microdialysis in the injured brain and include excitatory amino acids, ATP utilization (i.e., adenosine, inosine, hypoxantine) and depletion, neurotransmitters/neuromodulators release, cellular membrane degradation (glycerol) induction, and ROS generation [18]. Emphasizing the association between metabolic dysfunction, hypercatabolic states, and TBI severity contributes to some symptoms including fatigue and learning and memory dysfunctions [19]. Other factors that may transiently or persistently alter energy metabolism after TBI are decreased by cerebral blood flow (CBF) and cerebral ischemia, which may induce oxidative stress and, subsequently, mPTP opening [20].

Oxidative damage plays an important role in cell death pathway activation, axonal injury, and impaired synaptic plasticity, leading to cognitive deficits post-TBI [21,22]. Reactive oxygen species (ROS) generated by dysfunctional mitochondria and reactive nitrogen species generated through glutamate-mediated NMDA receptor activation can form the highly reactive species peroxynitrite ($ONOO^-$), which is toxic to cells. Peroxynitrite induces protein nitration, lipid peroxidation, and DNA fragmentation, activating caspase independent apoptotic cell death [23]. Any cellular constituent can be targeted by free radical damage, some of which increase cellular excitability after neuronal injury. Sodium-potassium pump (Na^+, K^+-ATPase) is one of these targets, being the main factor responsible for maintaining ion gradients across plasma membranes and controlling ATP consumption in neurons [24]. The inhibition of neuronal Na^+,K^+-ATPase activity with a concomitant increase of oxidative stress markers (protein carbonylation and lipoperoxidation) suggest that failure of this pump increases cellular excitability after TBI [25]. Furthermore, involvement of neutrophil infiltration, generation of inflammatory, and free radicals in the collapses of ion gradient homeostasis after neuronal injury has already been reported [26,27]. Accordingly, Na^+,K^+-ATPase inhibition is elicited by prostaglandin E2 [28], suggesting that the major prostaglandin lipid mediators of inflammation may increase brain excitability and, thus, contribute to a variety of TBI-induced inflammatory responses.

Frequently, TBI results in chronic disability characterized by multiple interacting secondary injury cascades. Considering that metabolic and functional deficits that occur after TBI are interrelated events, mechanistic studies may open new avenues to better understand TBI pathology and establish scientific-based rehabilitative strategies. There are currently few therapeutic options to mitigate the consequences of TBI, which is probably due to a single mechanism of action [29]. One of the possible strategies is the inclusion of ergogenic compounds, including caffeine, taurine, branch-chain amino acids, creatine, glutamine, and β-alanine. These supplements offer specific physiological advantages, which include enhanced energy availability/metabolism and improved recovery capacity [30,31]. Despite the well-recognized benefits of ergogenic aids in the form of dietary supplements on physical performance and/or recovery, these compounds were not used in this context in the present review [32]. Our goal herein is to summarize and discuss the potential prophylactic and/or therapeutic role of ergogenic compounds on the pathophysiology of TBI (Fig. 1).

3. Metabolic effect of creatine/phosphocreatine system in central nervous system

Creatine (N-[aminoiminomethyl]-*N*-methyl glycine) is a guanidine compound endogenously produced by the liver, kidneys, and pancreas from glycine, methionine, and arginine. Creatine is provided equally by the diet and endogenous synthesis by the rate-limiting enzyme L-arginine: glycine amidinotransferase (transamidinase), which conjugates arginine and glycine to produce guanidinoacetate. Then, guanidinoacetate is methylated by S-adenosyl-methionine and catalyzed by guanidinoacetate-methyltransferase (GAMT) to produce creatine [33]. The circulating creatine enters the cells via specific plasma membrane Na^+/Cl^- transporter, creatine transporter, and is transiently phosphorylated to phosphocreatine (PCr) by creatine kinases. Thus, when energetic needs are high, PCr can restitute the high energy phosphate bond to

Table 1
Summary of clinical and preclinical studies addressing the effect of ergogenic aid on pathophysiology induced by TBI.

Compound	Author	Dose	TBI model	Results
Creatine	Sakellaris et al., 2008 [52]	0.4 g/kg/day/6 months	Human	Duration of incubation reduction and post traumatic amnesia (PTA)
	Sullivan et al., 2000 [62]	0.1 mL/10 g body weight	CCI[a]	Protection against synaptic dysfunction and loss of cerebral cortical tissue
	Scheff and Dhillon, 2004 [63]	0.5%, 1% for 2 weeks before TBI	CCI	Lactate and free fatty acids diminishing and secondary injury processes markers increment related to mitochondrial integrity.
	Saraiva et al., 2012 [66]	300 mg/kg	FPI[b]	Antioxidant effect, protection against protein carbonylation and lipoperoxidation.
Carnitine	Scafidi et al., 2010 [93]	100 mg/kg	CCI	Protection against loss memory and reduce cortical lesion seven days after TBI in immature rats.
	Calikoglu et al., 2015 [95]	100 mg/kg in six times	WD[c]	Protection against inflammation and edema 72 h after TBI induction.
Arginine	DeWitt et al., 1997 [123]	100 mg/kg	FPI	Prevention of posttraumatic hypoperfusion.
	Wada et al., 1998 [129]	300 mg/kg	FPI	Protective effect by eNOS
	Cherian et al., 1999 [127]	300 mg/kg	CCI	Improvement of post-traumatic cerebral blood flow by increasing NO production.
	Cherian et al., 2003 [124]	300 mg/kg	CCI	Density of neuron in CA1 region increasing.
Taurine	Sun et al., 2015 [175]	2 (shows a tendency), 5, 15 and 50 mg/kg I.V.	WD	Reduction of oxidative stress and neuronal death in hippocampus, improvement of neurological functions.
	Su et al., 2014 [174]	200 mg/kg I.V. for 7 consecutive days	FPI	Reduced the expression of inflammatory cytokines, reactive astrogliosis, cerebral edema and improved neurological function following TBI.
	Wang et al., 2016 [173]	200 mg/kg I.V. for 7 consecutive days	FPI	Regulating coagulation and CBF state, reducing neuron damage in cortex and hippocampus and improving enzyme activity of mitochondria.
Glutamine	Feng, 2007 [229]	3% glutamine mixed powdered chow	WD	Decreased apoptosis and inflammatory markers in gut.
	Chen et al., 2008 [230]	3%glutamine mixed powdered chow	WD	Glutamine supplementation suppress the pro-inflammatory cytokines induced by TBI in the gut and reduced the intestinal apoptosis.
	Berg et al., 2006 [225]	0.34 g/kg	Human	Glutamine infusion increased plasma glutamine concentration by 30%, but not plasma glutamate concentration
Caffeine	Lusardi et al., 2012 [256]	25 mg/kg	FPI	Prevention of lethal apnea following fluid percussion injury and epileptiform bursting reduction.
	Li et al., 2008 [255]	(0.1 g/L, 0.25 g/L and 5.0 g/L)	WD	Glutamate release and inflammatory cytokine production attenuation are correlated with an upregulation of brain A_1 receptor mRNA.

[a] CCI, cortical contusion injury.
[b] FPI, fluid percussion injury.
[c] WD, weight drop.

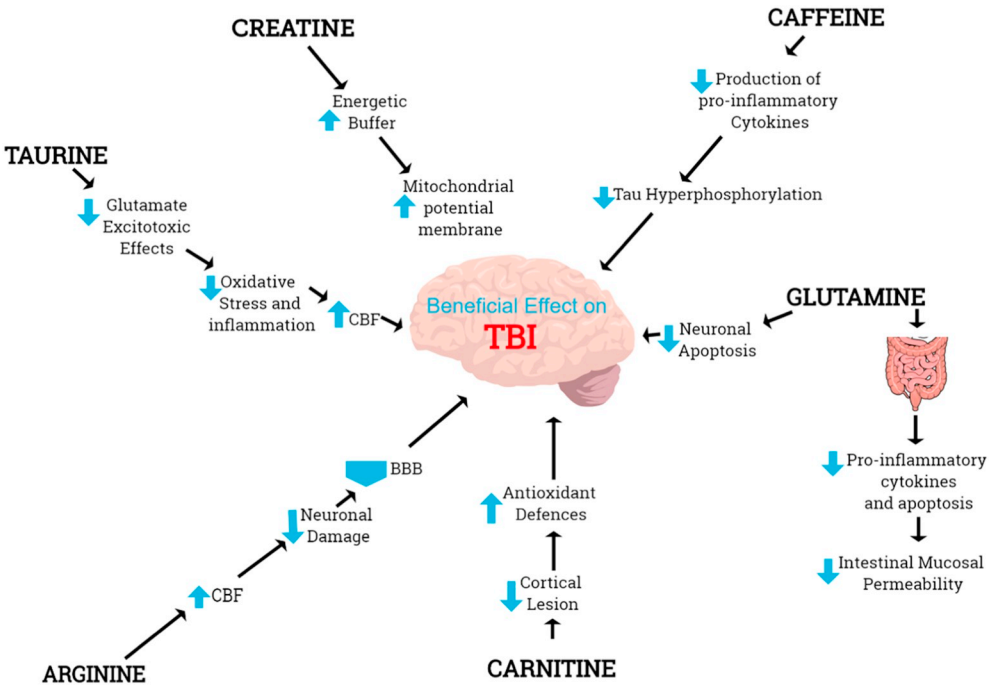

Fig. 1. Actions of ergogenic compounds in TBI prevention and/or treatment. In the schematic drawing above, the black arrows represent the beneficial effects of compost on the brain or gut, the blue up-arrows represent increases, the blue down-arrows represent decreases, and the blue shields represent protection. Creatine acts as an energetic buffer by keeping the mitochondria in operation, thus, it may contribute to diminished brain damage. Caffeine is commonly used in sports, although it decreases proinflammatory cytokines and contributes to minimizing Tau hyperphosphorylation in the brain. Glutamine decreases neuronal apoptosis in the brain, and in the gut, it decreases inflammatory processes and intestinal mucosal permeability. Carnitine contributes to increased antioxidant defenses and decreased cortical lesion, although little is known about its effects. L-arginine increases cerebral-blood-flow (CBF) and preserves the blood-brain-barrier by inhibiting matrix metalloproteinases, thus diminishing neuronal damage. Taurine decreases glutamate excitotoxicity effects, oxidative stress, and increases CBF. For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.

adenosine diphosphate (ADP) to regenerate ATP, which then becomes directly available for ATP-driven events [8].

Neurological deficits associated with inborn errors in creatine production and its storage reflect the importance of creatine to the central nervous system (CNS) [34]. Although creatine supplementation increases brain levels by one third as much as our muscular reserves, neuroprotection exerted by creatine includes enhanced energy buffering, which is a non-energetic role by rephosphorylating ADP that utilizes a proton (PCr + Mg ADP + H+ → creatine + Mg ATP) [35]. The ability to quench superoxide reinforces its protective effect due to antioxidant effects. Indeed, improved brain circulation [36] and specific binding of creatine to benzodiazepine binding sites of gamma aminobutyric acid type A ($GABA_A$) receptors [37] induced by creatine suggest neuroprotection. Moreover, PCr promotes glutamate uptake into synaptic vesicles [38], which may account for its neuroprotective capacity against glutamate toxicity in neuronal cell cultures [39].

Recently, a considerable body of evidence has suggested a non-energetic neuromodulatory role for creatine. Creatine is not only synthesized and taken up by neurons, but also released in an action-potential-dependent manner [40]. Furthermore, creatine represents a neuromodulator that may be released in the synapse, reuptaken by presynaptic plasma membrane creatine transporter (SLC6A8) and may either depress post-synaptic GABAergic neurotransmission or stimulate post-synaptic glutamatergic pathways. In this context, creatine increases the amplitude and number of population spikes in the stratum radiatum of the hippocampal CA1 subfield. This effect was attenuated by selective NMDA receptor antagonism. Creatine also increases [3H] MK-801 binding to hippocampal membranes by 55% [41] and leads to spatial learning improvement, which is possibly by modulating the binding site at the NMDA receptor [42] as well as molecular Ca^{2+}/calmodulin-dependent protein kinase II and cyclic AMP (cAMP)-response-element-binding protein phosphorylation [43]. Creatine has also been described as one of the main cellular osmolytes in muscles and CNS cells [44,45]. Indeed, in vitro studies involving rat cortical brain slices and primary astrocytic cultures using ^{1}H- and ^{31}P-magnetic resonance spectroscopy (MRS) have shown that decreased intracellular creatine promotes the opposite effect during hyper-osmotic conditions [46]. Decreased cerebral creatine was also demonstrated in an in vivo model of hepatic encephalopathy [47]. Finally, creatine has also been suggested as a potential regulator of the appetite and weight in hypothalamic nuclei [48–50].

Our current knowledge on creatine importance in normal neurological function and diseases is evidenced in a myriad of roles. It acts as potential sensor of cell methylation and energy status, a neuromodulator of GABAergic/glutamatergic pathway, a therapeutic agent endowed with anaplerotic properties, and bestows energetic and antioxidant actions along with osmolyte [5]. Creatine depletion in the brain may be associated with the disruption of neuronal functions and its augmentation in excitatory neurotransmitter efflux results in excitotoxicity after TBI. Therefore, it is useful to understand how supplemental creatine works to prevent or attenuate TBI-induced neurological and bioenergetic dysfunctions.

4. Creatine in TBI

Secondary damage caused by TBI is often larger in extent than the primary insult and is, at least in part, due to ischemia and cellular bioenergetic dysfunctions [9]. The reductions in *N*-acetyl aspartate (NAA)/Creatine, Choline (Cho)/Creatine, and/or NAA/Cho ratios observed in patients and animals with TBI [51] contribute to persistent symptoms after TBI. A prospective randomized study with children and adolescents suffering from TBI revealed that creatine supplementation improves clinical recovery in cognitive, personality/behavior, self-care, and communication aspects [52]. Indeed, daily creatine intake at a dosage of 0.4 g/kg in an oral suspension form for 6 months also decreased the duration of posttraumatic amnesia (PTA), intubation, and time in intensive care [53]. When patients experience rapid and acute brain or spinal cord injury it seems, however, unlikely that they immediately benefit from creatine supplementation, since exogenous creatine is absorbed slowly into CNS tissue [54]. These data suggest that this compound should be delivered as soon as possible after the insult directly to the injury sites, such as by perfusion of the affected region or intra-cerebroventricular administration, which are ways that have been shown to lead to quick increases of creatine levels in the

brain [9].

The ability of dietary creatine to alter brain energetics, promote neurogenesis, and improve brain function safely and effectively opens up the possibility for creatine monohydrate to provide a novel and natural strategy for treating TBI-induced psychiatric disorders [55,56]. The high-energy phosphoryl transfer reduction evidenced in TBI patients is not solely related to neuronal cell loss but may also depend on energetic impairment due to posttraumatic mitochondrial dysfunction [57,58]. In fact, perfusion-weighted imaging, three-dimensional (3D) magnetic resonance spectroscopic imaging, and diffusion tensor imaging (DTI) studies have detected many injuries in patients and animals with TBI characterized by impaired axonal function, neuronal metabolism, and perfusion [51,59,60].

Furthermore, experimental research has revealed that chronic ingestion of creatine before cortical damage increases brain levels of PCr [61] and ATP maintenance after moderate traumatic brain injury (moTBI) [62]. The same authors revealed that mitochondrial membrane potential significantly increased, intramitochondrial ROS and Ca^{2+} levels significantly decreased, and ATP levels were remained stable in animals fed with creatine [62]. These results are very interesting since reduced cellular ATP and increased ROS levels in synaptosomes may trigger neuronal cell death, and elevated intracellular Ca^{2+} levels trigger proteases and mPTP formation [16,62]. Moreover, creatine supplementation stabilizes mitochondrial CK activity after TBI, which is an enzyme associated to the mitochondrial adenine nucleotide translocator. This translocator inhibits mPTP through structural and functional interactions [62], an opening channel linked to mitochondrial depolarization and swelling, cessation of ATP synthesis, and apoptosis cascade [16]. The lactate and free fatty acid levels were also lower in animals treated with creatine before moTBI [63], suggesting that the mechanistic basis for the neuroprotective effects of creatine involves alterations of the insult-induced depletion of cellular ATP. Some evidence has pointed out interesting prophylactic properties of creatine, and the understanding of post-injury dysregulation is also essential to establish successful scientific-based strategies. One of these strategies may be supported by creatine, considering its role in reducing the initial damage, which limits secondary neuronal death outcomes and promotes neural repair and behavioral rehabilitation after TBI [5].

It is important to note that the progression of neuronal damage during the first 1–2 months post-injury corresponds to a period when most of the electrophysiology alterations imply in an increased excitability at injured tissue [64]. Recent data from our research group revealed that chronic changes in hippocampal GABAergic innervation are associated to seizure susceptibility induced by TBI and delayed supplementation (one week after neuronal injury) of creatine (300 mg/kg) protects against TBI-induced GABAergic dysfunction and reduced seizure susceptibility induced by subconvulsant pentylenetetrazole (PTZ), an effect that persists one week after creatine privation [65]. These experimental data reveal a great therapeutic window of opportunity for TBI treatment. Early creatine supplementation has been shown to protect against fluid percussion injury (FPI)-induced protein carbonylation and lipoperoxidation when neural analyses were performed 4- and 8-days post-injury [66]. However, creatine administration had no effect on Na^+,K^+-ATPase inhibition and TBI-induced seizures, which suggests that the antioxidant effects exerted by creatine do not protect against early excitatory inputs generated [66]. Furthermore, it is plausible to propose that protein carbonylation and lipoperoxidation occur separately from convulsive episodes and the ability of creatine to reduce this oxidative damage is not related to its anticonvulsant action. However, this discussion is speculative in nature and the mechanisms of action for creatine in certain biological systems and neurological diseases, such as TBI, have yet to be determined.

5. Metabolic effect of L-carnitine

L-Carnitine (β-hydroxy-y-trimethylaminobutyric acid) is a small water-soluble molecule involved in fatty acid metabolisms and physiologically synthesized by using L-lysine and l-methionine as substrates. L-Carnitine is present in red meat (e.g. beef and lamb) and other sources such as fish, poultry, and milk. Essentially, L-carnitine transports fatty acid chains into mitochondrial matrix, thus allowing cells to break down fat and acquire energy from stored fat reserves [67]. L-Carnitine is also found in plasma and tissue as free carnitine or bound to fatty acids as acylcarnitine derivatives [68]. In this context, the intracellular homeostasis of carnitine is controlled by different membrane transporters – the organic cation transporters (OCTNs), which operate on intestinal absorption and renal reabsorption of L-carnitine and play a major role in tissue distribution and variations in transport rates.

In mammals, L-carnitine is synthesized mainly in the liver, kidneys, and in some species by the testis and brain, from essential amino acids lysine and methionine as ultimate precursors to form trimethyllysine (TML) [69]. The availability of the intermediate TML limits carnitine biosynthesis, and most TML is located in skeletal muscles. Consequently, skeletal muscle protein turnover is considered the rate-limiting step of carnitine biosynthesis [70]. In CNS, acetyl-L-carnitine (ALC) can cross the blood-brain barrier (BBB) and act as a powerful antioxidant by preventing brain cell deterioration [71]. Moreover, ALC energizes the brain by improving the energy metabolism of neurons, equally facilitating the release and synthesis of neurotransmitters involved in short-term and long-term memory, such as acetylcholine [72]. Despite > 95% of the total carnitine in the body being stored within skeletal muscle tissues [73], ALC supplementation was demonstrated to modulate cerebral glucose utilization [74,75] and under some metabolic conditions, blood levels of free acetyl-CoA and ketones may become important for brain energy substrate [76]. Clinical and experimental trials with ALC showed that L-carnitine readily enters the brain and may delay the progression of Alzheimer's disease, relieve depression related to senility and other forms of dementia, and improve memory in the elderly [77–81].

6. L-Carnitine in TBI

Although mitochondria plays a pivotal role in cerebral metabolism and regulation of oxidative stress [82,83], TBI-induced cerebral metabolic crisis displays regional heterogeneity and varies temporally post-injury and according to injury severity [84,85]. In support of this perspective, TBI survivors show depletion of mitochondrial transcripts and proteins, reduced mitochondrial content, and cellular energetic failure, in addition to an early activation of the transcriptional program of mitochondrial biogenesis [86] In this regard, L-carnitine may improve mitochondrial function by detoxifying free-radicals and enhancing the activity of endogenous antioxidants [87,88]. In humans, a 7-day enteral feed with L-carnitine initiated within 24 h after severe TBI (sTBI) sustained both reduced brain edema and preserved neurological function for at least 90 days [89].

Evidence obtained from animal models of traumatic brain and spinal cord injury point towards the pyruvate dehydrogenase complex (PDHC) as a critical metabolic enzyme that is particularly sensitive to inhibition within a few hours after the event by either oxidative modifications or serine phosphorylation [90,91]. Considering that the overall reaction catalyzed by this complex constitutes the bridge between glycolysis and the first step of the tricarboxylic acid (TCA) cycle, impairment of the aerobic energy metabolism at the level of PDHC may potentially be overcome by using alternative fuels that enter the cycle distal. One of these fuels is ALC, a metabolic intermediate involved in transmitochondrial-membrane acetyl units trafficking for catabolic and anabolic metabolism inside the mitochondrial matrix that is converted into acetyl CoA and free carnitine via carnitine acyl transferases [92]. Consequently, ALC acts as a powerful antioxidant, which helps prevent brain cell deterioration in several TBI models [93–95] and patients suffering neurological injury [89,96].

In fact, [93] intraperitoneally (i.p.) ALC administration (100 mg/kg

in different times after neurological injury reduced cortical lesion by approximately 50% and protected against moTBI-induced memory dysfunction in immature rats using cortical controlled-impact (CCI) models. The neuronal acetyl CoA maintenance elicited by ALC administration protected against TBI-induced energy metabolism dysfunction and lactate accumulation [97,98]. It is important to note that exogenous ALC administration is also metabolized for energy via the TCA cycle in astrocytes and neurons within the forebrain [93]. In this sense, ALC incorporated into GABA, glutamate, and glutamine via TCA cycle metabolism protects against TBI-induced secondary damage [99]. In agreement to this view, previous studies have demonstrated the ability of this compound in activating antioxidant gene expressions [100] and general anti-inflammatory actions [101]. A previous study utilizing female Sprague-Dawley rats showed that L-Carnitine reduces oxidative stress and increases endogenous antioxidant MnSOD, with enhanced mitochondrial functional markers at 24 h after moderate brain injury with a small focal lesion [29]. In addition, since metabolic response to sTBI requires early nutritional resuscitation, plasma free carnitine decreases in TBI patients presenting 18 μmol/L median value, which reinforces the idea that carnitine and ester acetyl-L-carnitine utilization have neuro-protective effects [96].

7. Metabolic effect of L-arginine

L-arginine is a basic amino acid engaged in several mammalian metabolic pathways [102]. L-arginine is it not only a precursor for the synthesis of proteins, urea, and ornithine, but also polyamines, proline, glutamate, creatine, and agmatine [103]. Healthy adults synthesize adequate amounts of L-arginine and an important part comes directly from the diet [104]. L-arginine is used as a dietary supplement in subgroups of healthy individuals (i.e. bodybuilders, in whom standard supplemental doses are 12–18 g/day) and patients with all forms of malnutrition [105]. On the other hand, under conditions of stress, L-arginine supplementation must be acquired from the diet to maintain sufficient circulation and cellular L-arginine for essential bodily functions. In this context, approximately 10–12% of newborns exhibit L-arginine deficiency, a condition that inhibits normal growth and development [106]. Plasma L-arginine is also reported to be insufficient in low birth weight and preterm infants, as they are susceptible to intestinal inflammation and necrotizing enterocolitis [107].

Several factors affect the bioavailability of dietary L-arginine in infants and adults: the levels of lysine, manganese, and n-3 fatty acids in the diet and circulating hormones including cortisol, growth hormones, leptin, cytokines, endotoxins, as well as other biomolecules such as creatine, lactate, ornithine, and methylarginine [106]. L-arginine is the only substrate for nitric oxide (NO) production [108]. The NO is involved in several physiological processes including vasodilatation, cytotoxicity, immunity, and neurotransmission [109,110]. Considering that NO production induces vasodilation with a consequent elevation of blood flow in skeletal muscle during contraction, L-arginine supplementation reduces physiological fatigue by decreasing the plasma concentration of ammonia [108,111]. The L-arginine (1 g/kg) supplementation before a bout of eccentric exercise also improves the resolution of inflammation during the early phase of skeletal muscle repair and increases the expression of myogenic differentiation [112,113]. Moreover, NO inhibits platelet aggregation and neutrophil adhesion, in addition to scavenging superoxide generated during and after maximal or submaximal exercise [114]. Indeed, L-arginine has a protective role against oxidative stress, and this action is likely mediated via its interaction with superoxide ($O_2^{\cdot -}$) [115], inhibition of the xanthine oxidase pathway (an enzyme that produces ROS in rats subjected to hypoxia), and increased expression and/or activity of antioxidant enzymes [116–118].

8. L-Arginine in TBI

Regarding the effects of L-Arginine on TBI-induced pathophysiology, it is important to note that under physiological conditions, the amino group containing compounds exert fundamental biochemical roles in CNS [119]. For this reason, TBI studies have mainly focused on amino acid involvement in neurotransmission, especially regarding changes in their extracellular concentrations [120–122]. Considering that $O_2^{\cdot -}$ produced by TBI can inactivate NO by converting it to $ONOO^-$, NO maintenance elicited by L-arginine restores CBF, reduces neuronal damage, and protects against tyrosine nitration after TBI [123–126]. Moreover, NO production by neuronal and endothelial nitric oxide synthase (eNOS) also plays an obligatory role in CBF regulation, thus L-arginine-induced vasodilatation may decrease intracranial pressure after TBI [123,127–129]. L-arginine has intrinsic capabilities to cross the BBB and increment CBF without detrimentally increasing intracranial pressure [124,130,131]. Arginine-rich peptides also maintain mitochondrial integrity, reduce brain extracellular matrix destruction, and preserve BBB by inhibiting matrix metalloproteinases after TBI [132]. In this context, experimental research has shown that 300 mg/kg of L-arginine given within 5 min after neurological injury is the best dosage and therapeutic window to increase CBF [124,126,127,133] and neuron density in the hippocampal CA1 subfield [130], and reduce contusion volume [127,130], intracranial pressure [130] and nitrotyrosine-positive neurons in the cortex and hippocampus after TBI. Notably, contusion volume was only reduced when L-arginine was used 5 min or 1 h after TBI, although no adverse effects when it was administered 48 h after injury [130]. In humans, 300 mg/kg administered 12 and 24 h after sTBI did not change intracranial pressure or brain oxygenation, despite increasing internal carotid artery flow volume [134].

Many studies have suggested that the psychiatric, physical, and biological aspects of posttraumatic stress disorder (PTSD) may be associated with dysfunctions in several cellular processes including NO production [135–139]. Global arginine bioavailability, which is a marker of NO synthetic capacity in vivo, is lower in veterans with PTSD and negatively associated with some inflammation markers as well as with measures of PTSD symptom severity [140]. Reduced responsiveness to L-arginine after TBI may indicate that excessive damage to the cerebrovascular endothelium hinders its response and eNOS activity is inhibited, uncoupled, or L-arginine is diverted to another competing pathway, such as arginase. In line of this, arginase is the rate-limiting enzyme for ornithine production that serves as the precursor for proline and 4-hydroxyproline synthesis that form collagen [141–143]. Considering that TBI decrease collagen synthesis, it is plausible to propose that L-Arginine-induced collagen maintenance may be an important process of restoring BBB function after neuronal injury [141].

9. Metabolic effects of taurine

Taurine (2-aminoethane sulfonic acid) is an abundant free amino acid in the brain, retina, and skeletal muscle not used to synthesize proteins [144–146]. A small portion of taurine is produced in the liver and brain by two pathways. In the first way, it occurs from cysteine oxidation to cysteinesulfinic acid, is catalyzed by cysteine dioxygenase, decarboxylated by cysteine sulfonate decarboxylase, and oxidated by the resulting hypotaurine to taurine by a putative hypotaurine dehydrogenase [146]. In the second one, cysteine is incorporated into coenzyme A, followed by the release of cysteamine during coenzyme A turnover [147].

In the human body, taurine is absorbed from food throughout the small intestinal brush border membrane through Na^+ Cl^- dependent channels, such as taurine transporter (TauT) and the H^+/amino acid transporter 1 (PAT1) [146,148]. In recent years, energy drinks with a high content of taurine (1 g/250 mL) have become more frequent and present positive effects in the Peripheraland Central Nervous System

(CNS) through relaxation of pre-contracted airway smooth muscle cells in the lungs [149], and improvement of myocardial oxygen consumption, electrical activity, and exercise capacity in response to exercise in heart failure patients [150]. High taurine concentrations also increase Ca^{2+} accumulation rates in the sarcoplasmic reticulum and stimulate the process of excitation–contraction of skeletal muscles [151]. On the other hand, taurine depletion is associated with muscle cramping and exercise-induced muscle injury [152,153].

Peripheral taurine may be transported into the brain through the BBB by taurine transporters (TauT) [154,155]. Protective taurine effects occur through Ca^{2+} channel inhibition, such as Na^{+}/Ca^{2+} pumps, voltage-gated calcium channels (VGCCs), including L-, N-, and P/Q-types, and NMDA receptors [156–158]. Such inhibition reduces the glutamate-induced excitotoxicity [156,159] that activates the calpains, resulting in the inhibition of cytochrome C release and apoptosis cascade [156]. In addition, taurine influences proliferation signaling pathways of neural stem cells [160,161], acts as a mitochondrial modulator and protector [162–167], antioxidant [168–170], and can maintain cell membrane stability through the relation with Na^{+}/Ca^{2+} exchanger [156]. In addition to the beneficial effects as an ergogenic aid, taurine has potential therapeutic effects on several disorders, such as depression [154], diabetes mellitus [171], ischemic stroke [172], and TBI [173–175].

10. Taurine in TBI

It is well known that TBI alters the concentration of free amino acids in the CNS [119,176–180]. Considering taurine acts as an osmolyte in cell volume regulation [176], the extracellular taurine increase [176] in severely brain-injured patients [178] is attributed to a compensatory effect on glutamate-induced cell swelling [181–184]. In this manner, taurine interacts with NMDA receptor subtype, decreases the glutamate binding [156–158], and neutralizes glutamate excitotoxic effects after TBI. Recently, studies have supported the hypothesis that post-traumatic cytotoxic edema is directly related to mitochondrial function [185,186]. In this sense, taurine has been proven to improve the activity of respiratory chain complexes by depressing mitochondria-mediated cell death [187], removing free radicals such as $O_2^{\cdot -}$ and decreasing oxidative stress [173,188]. Considering that mitochondrial function is closely connected to glutamate toxicity, Ca^{2+} overload, and ROS production, a recent study concluded that taurine promotes an anti-glutamate toxicity effect by maintaining the normal mitochondria structure and respiratory complex activity [173].

In agreement to this view, Sun et al. [175] tested different doses (2, 5, 15, and 50 mg/kg) of taurine administered intravenously (i.v.) 30 min after TBI. The authors showed that taurine protects against body-weight loss and neurological dysfunctions characterized by oxidative stress and neuronal death in the hippocampus (CA1 and CA3 subfields) 7 days after TBI in a dose-dependent manner [189]. Similarly, taurine (50 mg/kg, i.v., 30 min and 4 h again after TBI) attenuates corpus callosum damage and hippocampal neuronal death (CA1 and CA3 subfields), in addition to suppressing the over-activation of calpain, which represents protective effects against grey and white matter damage [190]. However, other studies involving TBI have utilized an even higher dose of taurine (200/mg/kg) to attenuate or reverse TBI-induced dysfunction [173,174,191]. In this context, 200 mg/kg of taurine administered by tail i.v. injection once a day for 7 days after TBI, in addition to CBF, reduced neuron damage (cortex and hippocampus) and apoptosis rates, alleviated mitochondrial swelling, and increased the activities of respiratory complexes I and II [173]. Essentially, in all animal cells including astrocytes and neurons, the mechanism to adjust cell volume in response to osmolarity change is preserved to prevent disruption of the cytoarchitecture and maintain normal cell functions [192]. In order to re-establish their normal volume after swelling, astrocytes release taurine through a process termed regulatory volume decrease via volume-sensitive organic osmolyte/anion channel [193]. In TBI, such cell swelling is known as cytotoxic edema. Indeed, taurine concentration increases after TBI according to the duration of the cytotoxic edema observed morphologically in human patients [176,178,194,195].

11. Metabolic effects of glutamine

Circulating glutamine is the most abundant amino acid [196] and makes up > 20% of free amino acid pool in the blood and 40% in the muscles [197]. Glutamine is a crucial amino acid for the organism [198,199], and particularly under catabolic conditions, its consumption is augmented by the kidney, immune compartment, and gastrointestinal tract [200]. The diet can serve as a source of glutamine (a non-essential amino acid) from digested foods absorbed through the small intestine, although the human body can also synthesize it. Glutamine is maintained at a constant level in the circulation due to de novo synthesis and release from skeletal muscles, the lungs, and adipose tissues [201,202]. To maintain acid–base homeostasis, the kidneys release ammonia from glutamine, and nitrogen excess is eliminated by the liver and kidneys as urea from glutamine via the urea cycle [203].

Glutamine is absorbed into the cells by transporters [204] to be later used for biosynthesis or exported from cells by antiporters in exchange for other amino acids [205]. Glutamine is converted to ammonium ions and glutamate by mitochondrial glutaminases into cells. Thus, glutamate can be converted to α-ketoglutarate, which enters the TCA cycle to generate ATP through nicotinamide adenine dinucleotide and flavin adenine dinucleotide production. Glutamate can also contribute to tripeptide glutathione synthesis (glutamate-cysteine-glycine). In this regard, glutathione peroxidase neutralizes hydrogen peroxide, being the best known pathway by which glutamine controls ROS levels [206].

In physical exercises, for instance, glutamine supplementation is reported to occur during recovery after marathon running [207,208]. The ergogenic benefits are related to increased strength, quicker recovery, decreased infection frequency [209], prevention of overtraining [210], decreased proinflammatory cytokines [211], and intestinal permeability [212], along with increased anti-inflammatory cytokine levels [207,213]. Furthermore, glutamine increases heat shock protein expression [214,215] when associated with exercise [216]. Activation of the heat shock response leading to the inactivation of the nuclear factor-kappa B (NF-κB) inflammatory pathway may be the mechanism of cytokine inhibition provided by glutamine supplementation [217,218].

It is important to note that formulas containing elevated nutrient levels associated with immune modulation may positively influence immune response to stress in adult surgical and trauma patients [219]. Promising results from the feeding of enteral diets with specific additives, such as glutamine, arginine, omega-3 fatty acids, probiotics, symbiotics, and nucleotides have been reported recently, with various studies suggesting decreased septic complications, hospital costs or even mortality rates [220–222]. In this context, the understanding of how supplemental compounds, such as glutamine, may be applied to prevent or attenuate TBI-induced neurological dysfunction is extremely important.

12. Glutamine in TBI

Low plasma glutamine levels are associated with worse clinical outcomes, thus adequate nutrition may benefit patient care with TBI [223]. There is reluctance in using nutritional strategies through glutamine for TBI patients because it may result in increased intracerebral glutamate levels, which are associated to neuronal damage and apoptosis [224]. However, Berg and colleagues revealed that glutamine infusion (200 mg/mL for 20 h) following sTBI in humans increased plasma glutamine levels by 30%, although plasma glutamate concentrations remained unaltered [225]. In addition, Nägeli [226] showed that high doses of L-alanine-L-glutamine (0.75 g/kg via

continuous iv infusion for 5 days) increased plasma and brain glutamine and alanine levels, which were not associated with potential glutamate-mediated cerebral injury, thus suggesting positive effects of glutamine in TBI patients. However, the authors did not include only L-alanine or L-glutamine groups for more detailed comparisons [226]. Notably, this was the only study found to combine the selected ergogenic compounds considered in this review.

Traumatic brain injuries may also induce persistent inflammatory responses, several histopathological changes, and promote apoptosis in the intestine [227,228]. In this sense, TBI experimental studies have demonstrated that supplementation with glutamine (3% glutamine mixed powdered chow) 5 days after parietal cortical injury by the weight-dropping method decreased intestinal NF-κB activity, NF-kB p65 protein expression, concentrations of proinflammatory cytokines, and reduced apoptosis in the gut of male rats [229,230]. Indeed, Zhang [231] showed that glutamine supplementation protects against intestinal mucosal permeability and increased dopamine receptor expression after TBI. Fu et al. [232] evaluated the combination of hyperbaric oxygen treatment with glutamine nutritional therapy in TBI outcomes for controlled cortical impact and showed that this combination was effective in reducing neuronal apoptosis and improving neurological function in rats with TBI.

Clinical studies have investigated the enteral glutamine supplementation effect on immune function, wound healing, and length of hospital stays [233]. Glutamine supplementation after TBI normalizes the total lymphocyte and activated T helper lymphocyte counts, the response of lymphocytes to mitogens, as well as interleukin-2 plasma levels [234]. The effects of glutamine on TBI-induced damage may be an interesting target for therapeutic intervention for recovery and ameliorate functions in patients affected by this event.

13. Metabolic effects of caffeine

Caffeine (1,3,7-trimethyl-xanthine) is a purine alkaloid and considered the most widely consumed stimulant in the world [235]. About 80% of the world population consumes it daily and it is estimated that four out of five American individuals ingest it through food or drinks (coffee and tea) [236]. Caffeine is rapidly absorbed through the gastrointestinal tract in approximately 45 min, its plasma concentration peak is between 30 and 90 min, and its half-life is nearly 4–6 h [237,238]. This compound undergoes metabolism in the liver by the cytochrome P450 (CYP) enzyme CYP1A2. The enzymatic activity results in the primary metabolite paraxanthine (72–80%), however, theobromine and theophylline are also formed in smaller concentrations [239].

This stimulant compound exerts action in multiples pathways in the body. Caffeine may antagonize adenosine receptors (A1, A2A, A2B, and A3), being mostly mediated by the inhibition of high-affinity receptors A1 and A2A [240,241]. Many intracellular signaling pathways, including the cAMP pathway, affect these G-protein coupled receptors. The A1 and A3 subtype receptor activation leads to adenylate cyclase enzyme inhibition through an inhibitory G protein, decreasing intracellular levels of cAMP. On the other hand, the activation of A2A and A2B receptors stimulates adenylate cyclase of a stimulatory G protein by increasing the intracellular levels of cAMP [242]. In vascular smooth muscle cells, caffeine inhibits the cAMP degradating-enzyme (phosphodiesterase) by stimulating its intracellular signaling [243]. Caffeine also acts as a full agonist in ryanodine receptors and mobilizes Ca^2 ions from within the sarcoplasmic reticulum [241].

The pharmacological effects of caffeine include increased energy metabolism, decreased smooth muscle contraction, neurotransmitter release, and increased blood pressure [244]. Moreover, caffeine is one dietary component that has been extensively investigated for its role in sports. In fact, several reviews have documented the ergogenic properties of caffeine in endurance sports as well as in repeated or sustained high-intensity events [245,246]. In terms of endurance performance, caffeine may be ergogenic by sparing muscle glycogen, increasing force production, decreasing perceptions of pain, and stimulating the CNS, which delays fatigue and enhances athlete performance [247–249].

14. Caffeine in TBI

The ability of caffeine to ameliorate some neurodegenerative disorders has been proven in several studies, as it modulates many pathways in the organism [250]. Most studies discuss a relationship between the neuroprotective effect of caffeine and A2A receptor antagonism, which leads to decreased excitatory neurotransmitter release [251]. To determine the role of caffeine in TBI-induced pathophysiology, cerebrospinal fluid (CSF) concentrations of caffeine and its metabolites (theobromine, paraxanthine, and theophylline) were measured in adults after sTBI. Favorable Glasgow outcome scale was seen more often in patients with increased CSF caffeine concentrations, in addition to the fact that the caffeine metabolites theobromine and paraxanthine were also associated with favorable outcomes [252]. The authors speculate that caffeine may be neuroprotective by the long-term upregulation of adenosine A1 receptors or acute inhibition of A2A receptors [252].

Three weeks of caffeine administration in mice showed neuroprotection following a CCI characterized by neurological deficits, cerebral edema, and inflammatory cell infiltration (Li et al., 2008). The same study also revealed that chronic, but not acute caffeine administration, decreased glutamate release and inflammatory cytokine production, both effects correlated with an upregulation of brain A1 receptor mRNA after TBI. Furthermore, chronic caffeine consumption 3 weeks before TBI attenuates the decreased density and morphological degeneration of dendritic spines. Pharmacological A2 antagonism elicited by previous caffeine supplementation (0.25 g/L/day) reduced the level of tau phosphorylation and attenuated dendritic-spine degeneration, cognitive dysfunction, and spatial memory dysfunction induced by TBI [253], suggesting an interesting role of this ergogenic compound against TBI.

Nevertheless, many mechanisms involved in the evolution of secondary damage after TBI suggest that adverse and/or lack of protection of caffeine is associated with the dose and its effects through different pathways. For instance, caffeine at doses of 50, 100 or 150 mg/kg given i.p. 30 min before moTBI/sTBI increased righting latency, neurological deficiency, and mortality in a dose-dependent manner, in addition to exacerbating neutrophil infiltration, edema, disruption of the BBB, severe hemorrhage, and augmented lipid peroxidation [254]. This exacerbation of adverse outcomes after TBI is possibly a result of a blockade of adenosine receptors [254]. In another interesting study, 5 or 15 mg/kg of caffeine administered i.p. 30 min before TBI was ineffective in protecting against neurological deficits, apoptosis index, and glutamate and proinflammatory cytokine levels, while the dose of 50 mg/kg resulted in seizure activity and animal death [255]. However, 25 mg/kg of caffeine given i.p. right after sTBI prevented lethal apnea and epileptiform bursting reduction after sTBI, suggesting a potential role in modulating epilepsy development, although the compound did not change neuromotor function and histological outcomes [256]. Together, these studies with dose-response approaches in rodents indicate that a single dose of caffeine lower than 15 mg/kg or > 50 mg/kg is ineffective to counteract TBI and may even aggravate TBI-induced disturbances.

Ning et al. tested the use of caffeine (0.25 g/L/day) before (bTBI), after (aTBI), and before and after (baTBI) blast-induced TBI. Caffeine given before and/or after TBI improved memory deficit and protected against excitotoxicity, inflammation, astrogliosis, and neuronal loss at different stages of injury [257]. Notably, baTBI played positive a role in both early and prolonged stages of TBI, while bTBI and aTBI tended to exert neuroprotective effects (including adenosine A1R gene expression) at early and prolonged stages of TBI, respectively [257]. Afterwards, the same authors compared mice treated with caffeine (0.25 g/

L/day) for 29 days and withdrawal for one day before blast-induced TBI with the baTBI protocol explained above. The baTBI group showed alleviated brain injury, although the group also presented increased cumulative and time-segmented mortality postinjury [258]. However, more importantly, withdrawing caffeine intake before TBI resulted in favorable outcomes in mortality [258], although in vitro and in vivo animal studies suggest caffeine may also have a therapeutic role in patients with TBI, despite the risks and benefits of this ergogenic compound being controversial. Thus, long-term pre-clinical and clinical studies are necessary to better understand the action of caffeine in this neurological disease.

15. Conclusions

In summary, TBI is commonly followed by significant behavioral disabilities including permanent neurological disabilities and a wide range of behavioral and emotional problems. Clinical and experimental evidence suggests that the pervasive effects of TBI disrupt brain plasticity due to the inability of the brain to metabolize energy. In this context, a better understanding of neurological adaptations after this neuronal injury may provide important information necessary to develop new and more effective therapeutic strategies. One possible strategy may be the inclusion of ergogenic compounds, which is a popular term used for any device intended to enhance athletic performance, despite there being currently no FDA-approved therapeutic treatment for TBI. Although their functions are often unreported in clinical practice and poorly studied in animal models of TBI (many studies published in low-impact journals), promising results from ergogenic compounds suggest formulas containing nutrients that offer specific physiological advantages, including enhanced energy availability/metabolism and improved buffering capacity, which may positively influence the treatment of TBI-induced pathophysiology.

Declaration of Competing Interest

All authors declare no competing financial conflicts of interest.

Acknowledgments

The present work was supported by Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) (grants: 307382/2017-6). All authors have read and approved the final manuscript.

References

[1] M.C. Dewan, et al., Estimating the global incidence of traumatic brain injury, J. Neurosurg. (2018) 1–18.
[2] M. Faul, V. Coronado, Epidemiology of traumatic brain injury, Handb. Clin. Neurol. 127 (2015) 3–13.
[3] S. Binder, J.D. Corrigan, J.A. Langlois, The public health approach to traumatic brain injury: an overview of CDC's research and programs, J. Head Trauma Rehabil. 20 (3) (2005) 189–195.
[4] M. Majdan, et al., Epidemiology of traumatic brain injuries in Europe: a cross-sectional analysis, Lancet Public Health 1 (2) (2016) e76–e83.
[5] L.F. Freire Royes, G. Cassol, The effects of creatine supplementation and physical exercise on traumatic brain injury, Mini Rev Med Chem 16 (1) (2016) 29–39.
[6] T. King, et al., Life after the game—injury profile of past elite Australian football players, J. Sci. Med. Sport 16 (4) (2013) 302–306.
[7] J.G. Hobbs, J.S. Young, J.E. Bailes, Sports-related concussions: diagnosis, complications, and current management strategies, Neurosurg. Focus. 40 (4) (2016) E5.
[8] M. Wyss, R. Kaddurah-Daouk, Creatine and creatinine metabolism, Physiol. Rev. 80 (3) (2000) 1107–1213.
[9] R.H. Andres, et al., Functions and effects of creatine in the central nervous system, Brain Res. Bull. 76 (4) (2008) 329–343.
[10] K. Blennow, et al., Traumatic brain injuries, Nat Rev Dis Primers 2 (2016) 16084.
[11] K.R. Walker, G. Tesco, Molecular mechanisms of cognitive dysfunction following traumatic brain injury, Front. Aging Neurosci. 5 (2013) 29.
[12] D.A. Hovda, et al., The increase in local cerebral glucose utilization following fluid percussion brain injury is prevented with kynurenic acid and is associated with an increase in calcium, Acta Neurochir Suppl (Wien) 51 (1990) 331–333.
[13] G.A. Brooks, N.A. Martin, Cerebral metabolism following traumatic brain injury: new discoveries with implications for treatment, Front. Neurosci. 8 (2014) 408.
[14] M.J. McGinn, J.T. Povlishock, Cellular and molecular mechanisms of injury and spontaneous recovery, Handb. Clin. Neurol. 127 (2015) 67–87.
[15] L.K. Gilmer, et al., Early mitochondrial dysfunction after cortical contusion injury, J. Neurotrauma 26 (8) (2009) 1271–1280.
[16] G. Cheng, et al., Mitochondria in traumatic brain injury and mitochondrial-targeted multipotential therapeutic strategies, Br. J. Pharmacol. 167 (4) (2012) 699–719.
[17] P. Bouzat, P.J. Magistretti, M. Oddo, Hypertonic lactate and the injured brain: facts and the potential for positive clinical implications, Intensive Care Med. 40 (6) (2014) 920–921.
[18] P.J. Hutchinson, et al., Consensus statement from the 2014 International Microdialysis Forum, Intensive Care Med. 41 (9) (2015) 1517–1528.
[19] R. Vagnozzi, et al., Hypothesis of the postconcussive vulnerable brain: experimental evidence of its metabolic occurrence, Neurosurgery 57 (1) (2005) 164–171 (discussion 164-71).
[20] A.T. Mazzeo, et al., The role of mitochondrial transition pore, and its modulation, in traumatic brain injury and delayed neurodegeneration after TBI, Exp. Neurol. 218 (2) (2009) 363–370.
[21] A. Wu, Z. Ying, F. Gomez-Pinilla, Dietary curcumin counteracts the outcome of traumatic brain injury on oxidative stress, synaptic plasticity, and cognition, Exp. Neurol. 197 (2) (2006) 309–317.
[22] M. Ohta, et al., Attenuation of axonal injury and oxidative stress by edaravone protects against cognitive impairments after traumatic brain injury, Brain Res. 1490 (2013) 184–192.
[23] M.A. Ansari, K.N. Roberts, S.W. Scheff, A time course of contusion-induced oxidative stress and synaptic proteins in cortex in a rat model of TBI, J. Neurotrauma 25 (5) (2008) 513–526.
[24] Ames, A., 3rd, CNS energy metabolism as related to function. Brain Res. Brain Res. Rev., 2000. 34(1–2): p. 42–68.
[25] L.F. Silva, et al., Treadmill exercise protects against pentylenetetrazol-induced seizures and oxidative stress after traumatic brain injury, J. Neurotrauma 30 (14) (2013) 1278–1287.
[26] F.D. Lima, et al., Na^+,K^+-ATPase activity impairment after experimental traumatic brain injury: relationship to spatial learning deficits and oxidative stress, Behav. Brain Res. 193 (2) (2008) 306–310.
[27] B.C. Mota, et al., Exercise pre-conditioning reduces brain inflammation and protects against toxicity induced by traumatic brain injury: behavioral and neurochemical approach, Neurotox. Res. 21 (2) (2012) 175–184.
[28] M.S. Oliveira, et al., Prostaglandin E2 modulates Na^+,K^+-ATPase activity in rat hippocampus: implications for neurological diseases, J. Neurochem. 109 (2) (2009) 416–426.
[29] H. Chen, et al., L-carnitine and extendin-4 improve outcomes following moderate brain contusion injury, Sci. Rep. 8 (2018).
[30] R.M. Vogel, et al., Safety of a dose-escalated pre-workout supplement in recreationally active females, J Int Soc Sports Nutr 12 (2015) 12.
[31] K. De Pauw, et al., Do glucose and caffeine nasal sprays influence exercise or cognitive performance? Int J Sports Physiol Perform 12 (9) (2017) 1186–1191.
[32] M. Cameron, et al., The acute effects of a multi-ingredient pre-workout supplement on resting energy expenditure and exercise performance in recreationally active females, J Int Soc Sports Nutr 15 (1) (2018).
[33] M.A. Tarnopolsky, M.F. Beal, Potential for creatine and other therapies targeting cellular energy dysfunction in neurological disorders, Ann. Neurol. 49 (5) (2001) 561–574.
[34] M. Joncquel-Chevalier Curt, et al., Creatine biosynthesis and transport in health and disease, Biochimie 119 (2015) 146–165.
[35] T. Wallimann, M. Tokarska-Schlattner, U. Schlattner, The creatine kinase system and pleiotropic effects of creatine, Amino Acids 40 (5) (2011) 1271–1296.
[36] D. McGuire, et al., Localization of L-arginine-glycine amidinotransferase protein in rat tissues by immunofluorescence microscopy, Journal of Histochemistry & Cytochemistry 34 (4) (1986) 429–435.
[37] U. Dydak, et al., In vivo measurement of brain GABA concentrations by magnetic resonance spectroscopy in smelters occupationally exposed to manganese, Environ. Health Perspect. 119 (2) (2011) 219–224.
[38] O. Braissant, Creatine and guanidinoacetate transport at blood-brain and blood-cerebrospinal fluid barriers, J. Inherit. Metab. Dis. 35 (4) (2012) 655–664.
[39] O. Braissant, et al., Ammonium alters creatine transport and synthesis in a 3D culture of developing brain cells, resulting in secondary cerebral creatine deficiency, Eur. J. Neurosci. 27 (7) (2008) 1673–1685.
[40] L.S. Almeida, et al., Exocytotic release of creatine in rat brain, Synapse 60 (2) (2006) 118–123.
[41] L.F. Royes, et al., Neuromodulatory effect of creatine on extracellular action potentials in rat hippocampus: role of NMDA receptors, Neurochem. Int. 53 (1–2) (2008) 33–37.
[42] M.S. Oliveira, et al., The involvement of the polyamines binding sites at the NMDA receptor in creatine-induced spatial learning enhancement, Behav. Brain Res. 187 (1) (2008) 200–204.
[43] M.A. Souza, et al., Involvement of hippocampal CAMKII/CREB signaling in the spatial memory retention induced by creatine, Amino Acids 43 (6) (2012) 2491–2503.
[44] J.H. Bothwell, et al., Hypo-osmotic swelling-activated release of organic osmolytes in brain slices: implications for brain oedema in vivo, J. Neurochem. 77 (6) (2001) 1632–1640.
[45] R.R. Alfieri, et al., Creatine as a compatible osmolyte in muscle cells exposed to hypertonic stress, J. Physiol. 576 (Pt 2) (2006) 391–401.
[46] J.H. Bothwell, P. Styles, K.K. Bhakoo, Swelling-activated taurine and creatine

effluxes from rat cortical astrocytes are pharmacologically distinct, J. Membr. Biol. 185 (2) (2002) 157–164.
[47] O. Braissant, V.A. McLin, C. Cudalbu, Ammonia toxicity to the brain, J. Inherit. Metab. Dis. 36 (4) (2013) 595–612.
[48] R.B. Ceddia, G. Sweeney, Creatine supplementation increases glucose oxidation and AMPK phosphorylation and reduces lactate production in L6 rat skeletal muscle cells, J. Physiol. 555 (Pt 2) (2004) 409–421.
[49] R.A. Galbraith, M. Furukawa, M. Li, Possible role of creatine concentrations in the brain in regulating appetite and weight, Brain Res. 1101 (1) (2006) 85–91.
[50] J.D. Schmitto, et al., Miniaturized HIA microdiagonal pump as left ventricular assist device in a sheep model, ASAIO J. 54 (3) (2008) 233–236.
[51] B.L. Bartnik-Olson, et al., Impaired neurovascular unit function contributes to persistent symptoms after concussion: a pilot study, J. Neurotrauma 31 (17) (2014) 1497–1506.
[52] G. Sakellaris, et al., Prevention of complications related to traumatic brain injury in children and adolescents with creatine administration: an open label randomized pilot study, J. Trauma 61 (2) (2006) 322–329.
[53] G. Sakellaris, et al., Prevention of traumatic headache, dizziness and fatigue with creatine administration. A pilot study, Acta Paediatr. 97 (1) (2008) 31–34.
[54] O.S. Ipsiroglu, et al., Changes of tissue creatine concentrations upon oral supplementation of creatine-monohydrate in various animal species, Life Sci. 69 (15) (2001) 1805–1815.
[55] P.J. Allen, et al., Chronic creatine supplementation alters depression-like behavior in rodents in a sex-dependent manner, Neuropsychopharmacology 35 (2) (2010) 534–546.
[56] M.P. Cunha, et al., Evidence for the involvement of 5-HT1A receptor in the acute antidepressant-like effect of creatine in mice, Brain Res. Bull. 95 (2013) 61–69.
[57] S. Signoretti, et al., Assessment of mitochondrial impairment in traumatic brain injury using high-resolution proton magnetic resonance spectroscopy, J. Neurosurg. 108 (1) (2008) 42–52.
[58] R.A. Yeo, et al., A longitudinal proton magnetic resonance spectroscopy study of mild traumatic brain injury, J. Neurotrauma 28 (1) (2011) 1–11.
[59] I.I. Kirov, et al., Proton MR spectroscopy correlates diffuse axonal abnormalities with post-concussive symptoms in mild traumatic brain injury, J. Neurotrauma 30 (13) (2013) 1200–1204.
[60] V. Govind, et al., Whole-brain proton MR spectroscopic imaging of mild-to-moderate traumatic brain injury and correlation with neuropsychological deficits, J. Neurotrauma 27 (3) (2010) 483–496.
[61] P. Klivenyi, et al., Neuroprotective effects of creatine in a transgenic animal model of amyotrophic lateral sclerosis, Nat. Med. 5 (3) (1999) 347–350.
[62] P.G. Sullivan, et al., Dietary supplement creatine protects against traumatic brain injury, Ann. Neurol. 48 (5) (2000) 723–729.
[63] S.W. Scheff, H.S. Dhillon, Creatine-enhanced diet alters levels of lactate and free fatty acids after experimental brain injury, Neurochem. Res. 29 (2) (2004) 469–479.
[64] A. Pitkanen, K. Lukasiuk, Molecular and cellular basis of epileptogenesis in symptomatic epilepsy, Epilepsy Behav. 14 (Suppl. 1) (2009) 16–25.
[65] R.R. Gerbatin, et al., Delayed creatine supplementation counteracts reduction of GABAergic function and protects against seizures susceptibility after traumatic brain injury in rats, Prog. Neuro-Psychopharmacol. Biol. Psychiatry 92 (2019) 328–338.
[66] A.L. Saraiva, et al., Creatine reduces oxidative stress markers but does not protect against seizure susceptibility after severe traumatic brain injury, Brain Res. Bull. 87 (2–3) (2012) 180–186.
[67] J. Bremer, Carnitine—metabolism and functions, Physiol. Rev. 63 (4) (1983) 1420–1480.
[68] J.H. Walter, L-carnitine, Arch. Dis. Child. 74 (6) (1996) 475–478.
[69] F.M. Vaz, R.J. Wanders, Carnitine biosynthesis in mammals, Biochem. J. 361 (2002) 417–429 Pt 3.
[70] J. Pekala, et al., L-carnitine—metabolic functions and meaning in humans life, Curr. Drug Metab. 12 (7) (2011) 667–678.
[71] E. Vicari, A.E. Calogero, Effects of treatment with carnitines in infertile patients with prostato-vesiculo-epididymitis, Hum. Reprod. 16 (11) (2001) 2338–2342.
[72] G.C. Ferreira, M.C. McKenna, L-carnitine and acetyl-L-carnitine roles and neuroprotection in developing brain, Neurochem. Res. 42 (6) (2017) 1661–1675.
[73] E.P. Brass, Pharmacokinetic considerations for the therapeutic use of carnitine in hemodialysis patients, Clin. Ther. 17 (2) (1995) 176–185 (discussion 175).
[74] T. Aureli, et al., Acetyl-L-carnitine modulates glucose metabolism and stimulates glycogen synthesis in rat brain, Brain Res. 796 (1–2) (1998) 75–81.
[75] O.B. Smeland, et al., Chronic acetyl-L-carnitine alters brain energy metabolism and increases noradrenaline and serotonin content in healthy mice, Neurochem. Int. 61 (1) (2012) 100–107.
[76] Z. Binienda, et al., Neuroprotective effect of L-carnitine in the 3-nitropropionic acid (3-NPA)-evoked neurotoxicity in rats, Neurosci. Lett. 367 (2) (2004) 264–267.
[77] S. Berardi, et al., Characterization of L-carnitine transport into rat skeletal muscle plasma membrane vesicles, Eur. J. Biochem. 267 (7) (2000) 1985–1994.
[78] P. Zhou, et al., Acetyl-L-carnitine attenuates homocysteine-induced Alzheimer-like histopathological and behavioral abnormalities, Rejuvenation Res. 14 (6) (2011) 669–679.
[79] L.P. Klosinski, et al., White matter lipids as a ketogenic fuel supply in aging female brain: implications for Alzheimer's disease, EBioMedicine 2 (12) (2015) 1888–1904.
[80] A. Cristofano, et al., Serum levels of acyl-carnitines along the continuum from normal to Alzheimer's dementia, PLoS One 11 (5) (2016) e0155694.
[81] D. Ciavardelli, et al., Medium-chain plasma acylcarnitines, ketone levels, cognition, and gray matter volumes in healthy elderly, mildly cognitively impaired, or Alzheimer's disease subjects, Neurobiol. Aging 43 (2016) 1–12.
[82] J. Lifshitz, et al., Structural and functional damage sustained by mitochondria after traumatic brain injury in the rat: evidence for differentially sensitive populations in the cortex and hippocampus, J. Cereb. Blood Flow Metab. 23 (2) (2003) 219–231.
[83] C.L. Robertson, Mitochondrial dysfunction contributes to cell death following traumatic brain injury in adult and immature animals, J. Bioenerg. Biomembr. 36 (4) (2004) 363–368.
[84] D.K. Ragan, et al., Alterations in cerebral oxygen metabolism after traumatic brain injury in children, J. Cereb. Blood Flow Metab. 33 (1) (2013) 48–52.
[85] C.L. Robertson, et al., Mitochondrial mechanisms of cell death and neuroprotection in pediatric ischemic and traumatic brain injury, Exp. Neurol. 218 (2) (2009) 371–380.
[86] J.E. Carre, et al., Survival in critical illness is associated with early activation of mitochondrial biogenesis, Am. J. Respir. Crit. Care Med. 182 (6) (2010) 745–751.
[87] H. Chen, et al., L-carnitine and extendin-4 improve outcomes following moderate brain contusion injury, Sci. Rep. 8 (1) (2018) 11201.
[88] R. Zhang, et al., Neuroprotective effects of pre-treatment with l-carnitine and acetyl-L-carnitine on ischemic injury in vivo and in vitro, Int. J. Mol. Sci. 13 (2) (2012) 2078–2090.
[89] A. Mahmoodpoor, et al., A pilot trial of l-carnitine in patients with traumatic brain injury: effects on biomarkers of injury, J. Crit. Care 45 (2018) 128–132.
[90] C.L. Robertson, M. Saraswati, G. Fiskum, Mitochondrial dysfunction early after traumatic brain injury in immature rats, J. Neurochem. 101 (5) (2007) 1248–1257.
[91] U.W. Thomale, et al., Heterogeneous regional and temporal energetic impairment following controlled cortical impact injury in rats, Neurol. Res. 29 (6) (2007) 594–603.
[92] D.G. Cotter, R.C. Schugar, P.A. Crawford, Ketone body metabolism and cardiovascular disease, Am. J. Phys. Heart Circ. Phys. 304 (8) (2013) H1060–H1076.
[93] S. Scafidi, et al., Neuroprotection by acetyl-L-carnitine after traumatic injury to the immature rat brain, Dev. Neurosci. 32 (5–6) (2010) 480–487.
[94] M.L. Prins, J.H. Matsumoto, The collective therapeutic potential of cerebral ketone metabolism in traumatic brain injury, J. Lipid Res. 55 (12) (2014) 2450–2457.
[95] C. Calikoglu, et al., Effect of L-carnitine in preventing secondary damage in traumatic brain injury: an experimental study, Acta Medica Mediterranea 31 (4) (2015) 777–784.
[96] F. Vardon Bounes, et al., Plasma free carnitine in severe trauma: influence of the association with traumatic brain injury, Injury 49 (3) (2018) 538–542.
[97] M.L. Prins, et al., Increased cerebral uptake and oxidation of exogenous betaHB improves ATP following traumatic brain injury in adult rats, J. Neurochem. 90 (3) (2004) 666–672.
[98] A.A. Al-Majed, et al., Carnitine esters prevent oxidative stress damage and energy depletion following transient forebrain ischaemia in the rat hippocampus, Clin. Exp. Pharmacol. Physiol. 33 (8) (2006) 725–733.
[99] C. Vonder Haar, et al., Vitamins and nutrients as primary treatments in experimental brain injury: clinical implications for nutraceutical therapies, Brain Res. 1640 (Pt A) (2016) 114–129.
[100] S. Li, et al., L-carnitine protects renal tubular cells against calcium oxalate monohydrate crystals adhesion through preventing cells from dedifferentiation, Kidney Blood Press Res 41 (5) (2016) 582–592.
[101] C.P. Mescka, et al., Investigation of inflammatory profile in MSUD patients: benefit of L-carnitine supplementation, Metab. Brain Dis. 30 (5) (2015) 1167–1174.
[102] M.Z. Gad, Anti-aging effects of L-arginine, J. Adv. Res. 1 (3) (2010) 169–177.
[103] Morris, S.M., Jr., Arginine: beyond protein. Am. J. Clin. Nutr., 2006. 83(2): p. 508S–512S.
[104] M. Virarkar, et al., L-arginine and nitric oxide in CNS function and neurodegenerative diseases, Crit. Rev. Food Sci. Nutr. 53 (11) (2013) 1157–1167.
[105] L. Cynober, Pharmacokinetics of arginine and related amino acids, J. Nutr. 137 (6 Suppl 2) (2007) 1646S–1649S.
[106] G. Wu, et al., Arginine metabolism and nutrition in growth, health and disease, Amino Acids 37 (1) (2009) 153–168.
[107] P. Shah, V. Shah, Arginine supplementation for prevention of necrotising enterocolitis in preterm infants, Cochrane Database Syst. Rev. 3 (2007) (p. CD004339).
[108] R. Bescos, et al., The effect of nitric-oxide-related supplements on human performance, Sports Med. 42 (2) (2012) 99–117.
[109] M.Z. Gad, et al., Assessment of serum levels of asymmetric dimethylarginine, symmetric dimethylarginine and L-arginine in coronary artery disease, Biomarkers 15 (8) (2010) 746–752.
[110] S.J. Pearson, S.R. Hussain, A review on the mechanisms of blood-flow restriction resistance training-induced muscle hypertrophy, Sports Med. 45 (2) (2015) 187–200.
[111] A. Schaefer, et al., L-arginine reduces exercise-induced increase in plasma lactate and ammonia, Int. J. Sports Med. 23 (6) (2002) 403–407.
[112] T. Sakurai, et al., Role of nitric oxide in muscle regeneration following eccentric muscle contractions in rat skeletal muscle, J. Physiol. Sci. 63 (4) (2013) 263–270.
[113] A.D. Seabra, et al., Local inhibition of nitrergic activity in tenotomized rats accelerates muscle regeneration by increasing fiber area and decreasing central core lesions, Braz. J. Med. Biol. Res. 50 (3) (2017) e5556.
[114] L.H. Camargo, et al., Involvement of N-methyl-D-aspartate glutamate receptor and nitric oxide in cardiovascular responses to dynamic exercise in rats, Eur. J. Pharmacol. 713 (1–3) (2013) 16–24.
[115] S.J. Bailey, et al., Acute L-arginine supplementation reduces the O2 cost of moderate-intensity exercise and enhances high-intensity exercise tolerance, J Appl

Physiol (1985) 109 (5) (2010) 1394–1403.
[116] C.G. Cote, et al., Regulation of intracellular xanthine oxidase by endothelial-derived nitric oxide, Am. J. Phys. 271 (5 Pt 1) (1996) L869–L874.
[117] H. Shi, et al., Manipulation of arginase expression modulates abiotic stress tolerance in Arabidopsis: effect on arginine metabolism and ROS accumulation, J. Exp. Bot. 64 (5) (2013) 1367–1379.
[118] L. Shan, et al., L-arginine supplementation improves antioxidant defenses through L-arginine/nitric oxide pathways in exercised rats, J Appl Physiol (1985) 115 (8) (2013) 1146–1155.
[119] A.M. Amorini, et al., Severity of experimental traumatic brain injury modulates changes in concentrations of cerebral free amino acids, J. Cell. Mol. Med. 21 (3) (2017) 530–542.
[120] R. Chamoun, et al., Role of extracellular glutamate measured by cerebral microdialysis in severe traumatic brain injury, J. Neurosurg. 113 (3) (2010) 564–570.
[121] K. Purins, et al., Brain tissue oxygen monitoring: a study of in vitro accuracy and stability of Neurovent-PTO and Licox sensors, Acta Neurochir. 152 (4) (2010) 681–688.
[122] R.M. Guerriero, C.C. Giza, A. Rotenberg, Glutamate and GABA imbalance following traumatic brain injury, Curr Neurol Neurosci Rep 15 (5) (2015) 27.
[123] D.S. DeWitt, et al., L-arginine and superoxide dismutase prevent or reverse cerebral hypoperfusion after fluid-percussion traumatic brain injury, J. Neurotrauma 14 (4) (1997) 223–233.
[124] L. Cherian, C.S. Robertson, L-arginine and free radical scavengers increase cerebral blood flow and brain tissue nitric oxide concentrations after controlled cortical impact injury in rats, J. Neurotrauma 20 (1) (2003) 77–85.
[125] M.A. Avila, et al., L-arginine decreases fluid-percussion injury-induced neuronal nitrotyrosine immunoreactivity in rats, J. Cereb. Blood Flow Metab. 28 (10) (2008) 1733–1741.
[126] H. Liu, J.C. Goodman, C.S. Robertson, The effects of L-arginine on cerebral hemodynamics after controlled cortical impact injury in the mouse, J. Neurotrauma 19 (3) (2002) 327–334.
[127] L. Cherian, et al., Cerebral hemodynamic effects of phenylephrine and L-arginine after cortical impact injury, Crit. Care Med. 27 (11) (1999) 2512–2517.
[128] E. Morikawa, et al., L-arginine infusion promotes nitric oxide-dependent vasodilation, increases regional cerebral blood flow, and reduces infarction volume in the rat, Stroke 25 (2) (1994) 429–435.
[129] K. Wada, et al., Role of nitric oxide in traumatic brain injury in the rat, J. Neurosurg. 89 (5) (1998) 807–818.
[130] L. Cherian, et al., Neuroprotective effects of L-arginine administration after cortical impact injury in rats: dose response and time window, J. Pharmacol. Exp. Ther. 304 (2) (2003) 617–623.
[131] Z. He, et al., L-arginine ameliorates cerebral blood flow and metabolism and decreases infarct volume in rats with cerebral ischemia, Brain Res. 699 (2) (1995) 208–213.
[132] L.S. Chiu, et al., The neuroprotective potential of arginine-rich peptides for the acute treatment of traumatic brain injury, Expert. Rev. Neurother. 16 (4) (2016) 361–363.
[133] D.R. Mendez, L. Cherian, C.S. Robertson, Laser Doppler flow and brain tissue PO2 after cortical impact injury complicated by secondary ischemia in rats treated with arginine, J. Trauma 57 (2) (2004) 244–250.
[134] L. Rangel-Castilla, et al., L-arginine reactivity in cerebral vessels after severe traumatic brain injury, Neurol. Res. 32 (10) (2010) 1033–1040.
[135] J. Bugajski, et al., Blockade of nitric oxide formation impairs adrenergic-induced ACTH and corticosterone secretion, J. Physiol. Pharmacol. 50 (2) (1999) 327–334.
[136] O.A. Harris, et al., Infratentorial traumatic brain hemorrhage: may outcome be predicted by initial GCS? J. Trauma 49 (6) (2000) 1076–1082.
[137] M.O. Lopez-Figueroa, et al., Nitric oxide in the stress axis, Histol. Histopathol. 13 (4) (1998) 1243–1252.
[138] J. Persoons, et al., Acute stress affects cytokines and nitric oxide production by alveolar macrophages differently, Am. J. Respir. Crit. Care Med. 152 (2) (1995) 619–624.
[139] C.B. Yeh, et al., Characteristics of acute stress symptoms and nitric oxide concentration in young rescue workers in Taiwan, Psychiatry Res. 112 (1) (2002) 59–68.
[140] F.S. Bersani, et al., Mitochondrial DNA copy number is reduced in male combat veterans with PTSD, Prog. Neuro-Psychopharmacol. Biol. Psychiatry 64 (2016) 10–17.
[141] C.B. Jeter, et al., Human traumatic brain injury alters circulating L-arginine and its metabolite levels: possible link to cerebral blood flow, extracellular matrix remodeling, and energy status, J. Neurotrauma 29 (1) (2012) 119–127.
[142] A. Muellner, et al., Microvascular basal lamina antigen loss after traumatic brain injury in the rat, J. Neurotrauma 20 (8) (2003) 745–754.
[143] G. Louin, et al., Plasma concentrations of arginine and related amino acids following traumatic brain injury: proline as a promising biomarker of brain damage severity, Nitric Oxide 17 (2) (2007) 91–97.
[144] H. Ripps, W. Shen, Review: taurine: a “very essential” amino acid. Mol Vis, 18 (2012), pp. 2673–2686.
[145] T. Ito, et al., Tissue taurine depletion alters metabolic response to exercise and reduces running capacity in mice, J. Amino Acids 2014 (2014) 964680.
[146] V. Vitvitsky, S.K. Garg, R. Banerjee, Taurine biosynthesis by neurons and astrocytes, J. Biol. Chem. 286 (37) (2011) 32002–32010.
[147] M.H. Stipanuk, et al., Insights into taurine synthesis and function based on studies with cysteine dioxygenase (CDO1) knockout mice, Adv. Exp. Med. Biol. 803 (2015) 29–39.
[148] C.M. Anderson, et al., Taurine uptake across the human intestinal brush-border membrane is via two transporters: H + -coupled PAT1 (SLC36A1) and Na + - and Cl (-)-dependent TauT (SLC6A6), J. Physiol. 587 (2009) 731–744 Pt 4.
[149] G. Gallos, et al., Targeting the restricted alpha-subunit repertoire of airway smooth muscle GABAA receptors augments airway smooth muscle relaxation, Am J Physiol Lung Cell Mol Physiol 302 (2) (2012) L248–L256.
[150] M. Ahmadian, V. Dabidi Roshan, E. Ashourpore, Taurine supplementation improves functional capacity, myocardial oxygen consumption, and electrical activity in heart failure, J Diet Suppl 14 (4) (2017) 422–432.
[151] J.M. Eckerson, et al., Acute ingestion of sugar-free red bull energy drink has no effect on upper body strength and muscular endurance in resistance trained men, J Strength Cond Res 27 (8) (2013) 2248–2254.
[152] D. Conte Camerino, et al., Taurine and skeletal muscle disorders, Neurochem. Res. 29 (1) (2004) 135–142.
[153] T. Miyazaki, et al., Optimal and effective oral dose of taurine to prolong exercise performance in rat, Amino Acids 27 (3–4) (2004) 291–298.
[154] G.F. Wu, et al., Antidepressant effect of taurine in chronic unpredictable mild stress-induced depressive rats, Sci. Rep. 7 (1) (2017) 4989.
[155] I. Tamai, et al., Na(+)- and Cl(-)-dependent transport of taurine at the blood-brain barrier, Biochem. Pharmacol. 50 (11) (1995) 1783–1793.
[156] J.Y. Wu, H. Prentice, Role of taurine in the central nervous system, J. Biomed. Sci. 17 (Suppl. 1) (2010) S1.
[157] W.Q. Chen, et al., Role of taurine in regulation of intracellular calcium level and neuroprotective function in cultured neurons, J. Neurosci. Res. 66 (4) (2001) 612–619.
[158] H. Wu, et al., Mode of action of taurine as a neuroprotector, Brain Res. 1038 (2) (2005) 123–131.
[159] C. Pan, et al., Beneficial effect of taurine on hypoxia- and glutamate-induced endoplasmic reticulum stress pathways in primary neuronal culture, Amino Acids 43 (2) (2012) 845–855.
[160] X.W. Li, H.Y. Gao, J. Liu, The role of taurine in improving neural stem cells proliferation and differentiation, Nutr. Neurosci. 20 (7) (2017) 409–415.
[161] M.C. Shivaraj, et al., Taurine induces proliferation of neural stem cells and synapse development in the developing mouse brain, PLoS One 7 (8) (2012) e42935.
[162] A. El Idrissi, E. Trenkner, Growth factors and taurine protect against excitotoxicity by stabilizing calcium homeostasis and energy metabolism, J. Neurosci. 19 (21) (1999) 9459–9468.
[163] U. Warskulat, et al., Chronic liver disease is triggered by taurine transporter knockout in the mouse, FASEB J. 20 (3) (2006) 574–576.
[164] A. El Idrissi, E. Trenkner, Taurine regulates mitochondrial calcium homeostasis, Adv. Exp. Med. Biol. 526 (2003) 527–536.
[165] A. El Idrissi, Taurine increases mitochondrial buffering of calcium: role in neuroprotection, Amino Acids 34 (2) (2008) 321–328.
[166] M. Palmi, et al., Potentiation of mitochondrial Ca^{2+} sequestration by taurine, Biochem. Pharmacol. 58 (7) (1999) 1123–1131.
[167] S.H. Hansen, et al., The important role of taurine in oxidative metabolism, Adv. Exp. Med. Biol. 583 (2006) 129–135.
[168] L.K. Oharomari, et al., Exercise training and taurine supplementation reduce oxidative stress and prevent endothelium dysfunction in rats fed a highly palatable diet, Life Sci. 139 (2015) 91–96.
[169] Z. Yildirim, et al., Effects of taurine in cellular responses to oxidative stress in young and middle-aged rat liver, Ann. N. Y. Acad. Sci. 1100 (2007) 553–561.
[170] H. Nonaka, et al., Taurine prevents the decrease in expression and secretion of extracellular superoxide dismutase induced by homocysteine: amelioration of homocysteine-induced endoplasmic reticulum stress by taurine, Circulation 104 (10) (2001) 1165–1170.
[171] F.L. Rahmeier, et al., The effect of taurine and enriched environment on behaviour, memory and hippocampus of diabetic rats, Neurosci. Lett. 630 (2016) 84–92.
[172] J. Menzie, H. Prentice, J.Y. Wu, Neuroprotective mechanisms of taurine against ischemic stroke, Brain Sci 3 (2) (2013) 877–907.
[173] Q. Wang, et al., Protective effects of taurine in traumatic brain injury via mitochondria and cerebral blood flow, Amino Acids 48 (9) (2016) 2169–2177.
[174] Y. Su, et al., Taurine improves functional and histological outcomes and reduces inflammation in traumatic brain injury, Neuroscience 266 (2014) 56–65.
[175] M. Sun, et al., Protective effects of taurine against closed head injury in rats, J. Neurotrauma 32 (1) (2015) 66–74.
[176] Y. Seki, et al., Cerebrospinal fluid taurine after traumatic brain injury, Neurochem. Res. 30 (1) (2005) 123–128.
[177] P. Nilsson, et al., Changes in cortical extracellular levels of energy-related metabolites and amino acids following concussive brain injury in rats, J. Cereb. Blood Flow Metab. 10 (5) (1990) 631–637.
[178] J.F. Stover, et al., Glutamate and taurine are increased in ventricular cerebrospinal fluid of severely brain-injured patients, J. Neurotrauma 16 (2) (1999) 135–142.
[179] J.M. Pascual, et al., Time course of early metabolic changes following diffuse traumatic brain injury in rats as detected by (1)H NMR spectroscopy, J. Neurotrauma 24 (6) (2007) 944–959.
[180] K. Singh, et al., Study of neurometabolic and behavioral alterations in rodent model of mild traumatic brain injury: a pilot study, NMR Biomed. 29 (12) (2016) 1748–1758.
[181] N. Menendez, et al., Extracellular taurine increase in rat hippocampus evoked by specific glutamate receptor activation is related to the excitatory potency of glutamate agonists, Neurosci. Lett. 102 (1) (1989) 64–69.
[182] G.R. Dutton, et al., Astrocyte taurine, Ann. N. Y. Acad. Sci. 633 (1991) 489–500.
[183] P. Saransaari, S.S. Oja, Excitatory amino acids evoke taurine release from cerebral cortex slices from adult and developing mice, Neuroscience 45 (2) (1991) 451–459.
[184] J.F. Stover, A.W. Unterberg, Increased cerebrospinal fluid glutamate and taurine

concentrations are associated with traumatic brain edema formation in rats, Brain Res. 875 (1–2) (2000) 51–55.
[185] A. Millet, et al., Hypertonic sodium lactate reverses brain oxygenation and metabolism dysfunction after traumatic brain injury, Br. J. Anaesth. 120 (6) (2018) 1295–1303.
[186] E. Vlodavsky, et al., Post-traumatic cytotoxic edema is directly related to mitochondrial function, J. Cereb. Blood Flow Metab. 37 (1) (2017) 166–177.
[187] M. Sun, et al., Protective functions of taurine against experimental stroke through depressing mitochondria-mediated cell death in rats, Amino Acids 40 (5) (2011) 1419–1429.
[188] Q. Sun, et al., Taurine attenuates amyloid beta 1-42-induced mitochondrial dysfunction by activating of SIRT1 in SK-N-SH cells, Biochem. Biophys. Res. Commun. 447 (3) (2014) 485–489.
[189] G. Aad, et al., Search for Higgs boson pair production in the gammagammabb [over] final state using pp collision data at sqrt[s] = 8 TeV from the ATLAS detector, Phys. Rev. Lett. 114 (8) (2015) 81802.
[190] Y. Gu, et al., Taurine attenuates hippocampal and corpus callosum damage, and enhances neurological recovery after closed head injury in rats, Neuroscience 291 (2015) 331–340.
[191] R. Gupte, et al., Evaluation of taurine neuroprotection in aged rats with traumatic brain injury, Brain Imaging Behav 13 (2) (2018) 461–471.
[192] H. Pasantes-Morales, O. Quesada, J. Moran, Taurine: an osmolyte in mammalian tissues, Adv. Exp. Med. Biol. 442 (1998) 209–217.
[193] H.K. Kimelberg, Receptors on astrocytes—what possible functions? Neurochem. Int. 26 (1) (1995) 27–40.
[194] R. Bullock, et al., Correlation of the extracellular glutamate concentration with extent of blood flow reduction after subdural hematoma in the rat, J. Neurosurg. 74 (5) (1991) 794–802.
[195] D.A.R. Mc, K. Opeskin, Timing of early changes in brain trauma, Am J Forensic Med Pathol 19 (1) (1998) 1–9.
[196] J.R. Mayers, M.G. Vander Heiden, Famine versus feast: understanding the metabolism of tumors in vivo, Trends Biochem. Sci. 40 (3) (2015) 130–140.
[197] J. Bergstrom, et al., Intracellular free amino acid concentration in human muscle tissue, J. Appl. Physiol. 36 (6) (1974) 693–697.
[198] Ceddia, M.A., E.W. Voss, Jr., and J.A. Woods, Intracellular mechanisms responsible for exercise-induced suppression of macrophage antigen presentation. J Appl Physiol (1985), 2000. 88(2): p. 804–10.
[199] P. Newsholme, et al., Glutamine and glutamate as vital metabolites, Braz. J. Med. Biol. Res. 36 (2) (2003) 153–163.
[200] J.M. Lacey, D.W. Wilmore, Is glutamine a conditionally essential amino acid? Nutr. Rev. 48 (8) (1990) 297–309.
[201] M. Stumvoll, et al., Role of glutamine in human carbohydrate metabolism in kidney and other tissues, Kidney Int. 55 (3) (1999) 778–792.
[202] C.T. Hensley, A.T. Wasti, R.J. DeBerardinis, Glutamine and cancer: cell biology, physiology, and clinical opportunities, J. Clin. Invest. 123 (9) (2013) 3678–3684.
[203] L. Taylor, N.P. Curthoys, Glutamine metabolism: role in acid-base balance*, Biochem. Mol. Biol. Educ. 32 (5) (2004) 291–304.
[204] Y.D. Bhutia, et al., Amino acid transporters in cancer and their relevance to "glutamine addiction": novel targets for the design of a new class of anticancer drugs, Cancer Res. 75 (9) (2015) 1782–1788.
[205] P. Nicklin, et al., Bidirectional transport of amino acids regulates mTOR and autophagy, Cell 136 (3) (2009) 521–534.
[206] T.C. Welbourne, Ammonia production and glutamine incorporation into glutathione in the functioning rat kidney, Can. J. Biochem. 57 (3) (1979) 233–237.
[207] L. Castell, Glutamine supplementation in vitro and in vivo, in exercise and in immunodepression, Sports Med. 33 (5) (2003) 323–345.
[208] L.M. Castell, E.A. Newsholme, The effects of oral glutamine supplementation on athletes after prolonged, exhaustive exercise, Nutrition 13 (7–8) (1997) 738–742.
[209] L.M. Castell, J.R. Poortmans, E.A. Newsholme, Does glutamine have a role in reducing infections in athletes? Eur. J. Appl. Physiol. Occup. Physiol. 73 (5) (1996) 488–490.
[210] G.C. Phillips, Glutamine: the nonessential amino acid for performance enhancement, Curr Sports Med Rep 6 (4) (2007) 265–268.
[211] V.F. Cruzat, M.M. Rogero, J. Tirapegui, Effects of supplementation with free glutamine and the dipeptide alanyl-glutamine on parameters of muscle damage and inflammation in rats submitted to prolonged exercise, Cell Biochem. Funct. 28 (1) (2010) 24–30.
[212] G.P. Lambert, et al., Gastrointestinal permeability during exercise: effects of aspirin and energy-containing beverages, J Appl Physiol (1985) 90 (6) (2001) 2075–2080.
[213] N. Hiscock, L.T. Mackinnon, A comparison of plasma glutamine concentration in athletes from different sports, Med. Sci. Sports Exerc. 30 (12) (1998) 1693–1696.
[214] G. Guiqi, Pre-treatment with glutamine attenuates lung injury in rats subjected to intestinal ischaemia-reperfusion, Injury 42 (1) (2011) 72–77.
[215] J.A. Ehrenfried, et al., Glutamine-mediated regulation of heat shock protein expression in intestinal cells, Surgery 118 (2) (1995) 352–356 (discussion 356-7).
[216] M. Zuhl, et al., The effects of acute oral glutamine supplementation on exercise-induced gastrointestinal permeability and heat shock protein expression in peripheral blood mononuclear cells, Cell Stress Chaperones 20 (1) (2015) 85–93.
[217] K. Dokladny, et al., LPS-induced cytokine levels are repressed by elevated expression of HSP70 in rats: possible role of NF-kappaB, Cell Stress Chaperones 15 (2) (2010) 153–163.
[218] T. Paimela, et al., Celastrol regulates innate immunity response via NF-kappaB and Hsp70 in human retinal pigment epithelial cells, Pharmacol. Res. 64 (5) (2011) 501–508.
[219] T.J. Painter, J. Rickerds, R.F. Alban, Immune enhancing nutrition in traumatic brain injury - a preliminary study, Int. J. Surg. 21 (2015) 70–74.
[220] U. Sonnewald, et al., Metabolism of [U-13C5] glutamine in cultured astrocytes studied by NMR spectroscopy: first evidence of astrocytic pyruvate recycling, J. Neurochem. 67 (6) (1996) 2566–2572.
[221] R.D. Griffiths, Outcome of critically ill patients after supplementation with glutamine, Nutrition 13 (7–8) (1997) 752–754.
[222] S.C. Lee-anne, et al., Nutrition therapy in the optimisation of health outcomes in adult patients with moderate to severe traumatic brain injury: findings from a scoping review, Injury 45 (12) (2014) 1834–1841.
[223] P.C. Rodas, et al., Glutamine and glutathione at ICU admission in relation to outcome, Clin Sci (Lond) 122 (12) (2012) 591–597.
[224] C. Zwingmann, D. Leibfritz, A.S. Hazell, Nmr spectroscopic analysis of regional brain energy metabolism in manganese neurotoxicity, Glia 55 (15) (2007) 1610–1617.
[225] A. Berg, et al., Intravenous glutamine supplementation to head trauma patients leaves cerebral glutamate concentration unaffected, Intensive Care Med. 32 (11) (2006) 1741–1746.
[226] M. Nageli, et al., Prolonged continuous intravenous infusion of the dipeptide L-alanine- L-glutamine significantly increases plasma glutamine and alanine without elevating brain glutamate in patients with severe traumatic brain injury, Crit. Care 18 (4) (2014) R139.
[227] P.L. Faries, et al., Intestinal permeability correlates with severity of injury in trauma patients, J. Trauma 44 (6) (1998) 1031–1035 (discussion 1035-6).
[228] C.H. Hang, et al., Expressions of intestinal NF-kappaB, TNF-alpha, and IL-6 following traumatic brain injury in rats, J. Surg. Res. 123 (2) (2005) 188–193.
[229] D. Feng, et al., Influence of glutamine on intestinal inflammatory response, mucosa structure alterations and apoptosis following traumatic brain injury in rats, J Int Med Res 35 (5) (2007) 644–656.
[230] G. Chen, et al., Glutamine decreases intestinal nuclear factor kappa B activity and pro-inflammatory cytokine expression after traumatic brain injury in rats, Inflamm. Res. 57 (2) (2008) 57–64.
[231] X. Zhang, X. Jiang, Effects of enteral nutrition on the barrier function of the intestinal mucosa and dopamine receptor expression in rats with traumatic brain injury, JPEN J. Parenter. Enteral Nutr. 39 (1) (2015) 114–123.
[232] X. Fu, et al., Hyperbaric oxygen treatment and enteral nutrition support with glutamine relieves traumatic brain injury in the rats, Int. J. Clin. Exp. Med. 7 (12) (2014) 5686–5690.
[233] L. Curtis, P. Epstein, Nutritional treatment for acute and chronic traumatic brain injury patients, J. Neurosurg. Sci. 58 (3) (2014) 151–160.
[234] K.J. Lorenz, R. Schallert, V. Daniel, Immunonutrition - the influence of early postoperative glutamine supplementation in enteral/parenteral nutrition on immune response, wound healing and length of hospital stay in multiple trauma patients and patients after extensive surgery, GMS Interdiscip Plast Reconstr Surg DGPW 4 (2015) Doc15.
[235] E. Harpaz, et al., The effect of caffeine on energy balance, J. Basic Clin. Physiol. Pharmacol. 28 (1) (2017) 1–10.
[236] B.F. Harland, Caffeine and nutrition, Nutrition 16 (7–8) (2000) 522–526.
[237] T.E. Graham, Caffeine and exercise: metabolism, endurance and performance, Sports Med. 31 (11) (2001) 785–807.
[238] C.E. Carvey, L.A. Thompson, C.R. Mahoney, Caffeine: mechanism of action, genetics, and behavioral studies conducted in task, Sleep Deprivation, Stimulant Medications, and Cognition, 2012, p. 93.
[239] M.A. Heckman, J. Weil, E. Gonzalez de Mejia, Caffeine (1, 3, 7-trimethylxanthine) in foods: a comprehensive review on consumption, functionality, safety, and regulatory matters, J. Food Sci. 75 (3) (2010) R77–R87.
[240] P. Nawrot, et al., Effects of caffeine on human health, Food Addit. Contam. 20 (1) (2003) 1–30.
[241] B.B. Fredholm, K. Lindstrom, Autoradiographic comparison of the potency of several structurally unrelated adenosine receptor antagonists at adenosine A1 and A(2A) receptors, Eur. J. Pharmacol. 380 (2–3) (1999) 197–202.
[242] M.E. Olah, G.L. Stiles, Adenosine receptor subtypes: characterization and therapeutic regulation, Annu. Rev. Pharmacol. Toxicol. 35 (1995) 581–606.
[243] S. Puttachary, et al., Levamisole and ryanodine receptors. II: an electrophysiological study in Ascaris suum, Mol. Biochem. Parasitol. 171 (1) (2010) 8–16.
[244] A. Nehlig, Interindividual differences in caffeine metabolism and factors driving caffeine consumption, Pharmacol. Rev. 70 (2) (2018) 384–411.
[245] M.S. Ganio, et al., Effect of caffeine on sport-specific endurance performance: a systematic review, J Strength Cond Res 23 (1) (2009) 315–324.
[246] T.A. Astorino, et al., Effect of two doses of caffeine on muscular function during isokinetic exercise, Med. Sci. Sports Exerc. 42 (12) (2010) 2205–2210.
[247] C.A. Bridge, M.A. Jones, The effect of caffeine ingestion on 8 km run performance in a field setting, J. Sports Sci. 24 (4) (2006) 433–439.
[248] R.E. Beaumont, L.J. James, Effect of a moderate caffeine dose on endurance cycle performance and thermoregulation during prolonged exercise in the heat, J. Sci. Med. Sport 20 (11) (2017) 1024–1028.
[249] J.S. Chia, et al., Effects of caffeine supplementation on performance in ball games, Sports Med. 47 (12) (2017) 2453–2471.
[250] J. Manchester, I. Eshel, D.W. Marion, The benefits and risks of energy drinks in young adults and military service members, Mil. Med. 182 (7) (2017) e1726–e1733.
[251] A. Kalda, et al., Novel neuroprotection by caffeine and adenosine A(2A) receptor antagonists in animal models of Parkinson's disease, J. Neurol. Sci. 248 (1–2) (2006) 9–15.
[252] K.T. Sachse, et al., Increases in cerebrospinal fluid caffeine concentration are associated with favorable outcome after severe traumatic brain injury in humans, J.

Cereb. Blood Flow Metab. 28 (2) (2008) 395–401.
[253] Z.A. Zhao, et al., Adenosine A2A receptor inactivation alleviates early-onset cognitive dysfunction after traumatic brain injury involving an inhibition of tau hyperphosphorylation, Transl. Psychiatry 7 (5) (2017) e1123.
[254] K. Al Moutaery, et al., Caffeine impairs short-term neurological outcome after concussive head injury in rats, Neurosurgery 53 (3) (2003) 704–711 (discussion 711-2).
[255] W. Li, et al., Chronic but not acute treatment with caffeine attenuates traumatic brain injury in the mouse cortical impact model, Neuroscience 151 (4) (2008) 1198–1207.
[256] T.A. Lusardi, et al., Caffeine prevents acute mortality after TBI in rats without increased morbidity, Exp. Neurol. 234 (1) (2012) 161–168.
[257] Y.L. Ning, et al., Chronic caffeine exposure attenuates blast-induced memory deficit in mice, Chin. J. Traumatol. 18 (4) (2015) 204–211.
[258] Y.L. Ning, et al., Caffeine attenuates brain injury but increases mortality induced by high-intensity blast wave exposure, Toxicol. Lett. 301 (2019) 90–97.