Am J Neurodegener Dis 2023;12(1):1-15
www.AJND.us /ISSN:2165-591X/AJND0147534

Original Article

Cognitive impairment and changes of red blood cell components and serum levels of IL-6, IL-18, and L-tryptophan in methamphetamine abusers

Nutthika Chaidee[1], Natcharee Kraiwattanapirom[1], Supitcha Pannengpetch[2], Chutikorn Nopparat[3], Piyarat Govitrapong[4], Vorasith Siripornpanich[1], Wilasinee Suwanjang[2], Sutisa Nudmamud-Thanoi[5], Banthit Chetsawang[1]

[1]Research Center for Neuroscience, Institute of Molecular Biosciences, Mahidol University, Salaya, Nakhon Pathom, Thailand; [2]Center for Research and Innovation, Faculty of Medical Technology, Mahidol University, Salaya, Nakhon Pathom, Thailand; [3]Innovative Learning Center, Srinakharinwirot University, Bangkok, Thailand; [4]Chulabhorn Graduate Institute, Chulabhorn Royal Academy, Bangkok, Thailand; [5]Department of Anatomy and Centre of Excellence in Medical Biotechnology, Faculty of Medical Science, Naresuan University, Phitsanulok, Thailand

Received November 4, 2022; Accepted February 8, 2023; Epub February 25, 2023; Published February 28, 2023

Abstract: The deficit in cognitive function is more concerning in methamphetamine (MA) users. The cognitive deficit was suspected to be the consequence of neuroinflammation-induced neurological dysregulation. In addition, activating the key enzyme in the tryptophan metabolic pathway by pro-inflammatory cytokines results in metabolite toxicity, further generating cognitive impairments. However, the evidence for the role of neuroinflammation and tryptophan metabolites involved in MA-induced cognitive deficit needs more conclusive study. Objectives: This retrospective study aimed to determine blood-inflammatory markers, tryptophan metabolite-related molecules, and cognitive function in MA abusers compared to healthy control (HC) participants. Methods: The cognitive functions were evaluated using Stroop, Go/No-Go, One Back Task (OBT), and Wisconsin Card Sorting Test-64 (WCST-64). Blood samples were analyzed for complete blood count (CBC) analysis, serum inflammatory cytokines interleukin (IL)-6 and IL-18 and tryptophan metabolites. Results: MA group exhibited poor cognitive performance in selective attention, inhibition, working memory, cognitive flexibility, concept formation and processing speed compared to HC. Reduction in red blood cell (RBC) components but induction in white blood cells (WBCs) and IL-6 were observed in MA abusers, which might indicate anemia of (systemic chronic low-grade) inflammation. In addition, the depletion of precursor in the tryptophan metabolic pathway, L-tryptophan was also observed in MA users, which might represent induction in tryptophan metabolites. Conclusion: These findings emphasize that blood biomarkers might be a surrogate marker to predict the role of neuroinflammation and abnormal tryptophan metabolite in MA-induced cognitive impairments.

Keywords: Methamphetamine, cognitive impairments, inflammation, tryptophan metabolites, anemia of inflammation, biomarkers

Introduction

Recent evidence demonstrated that methamphetamine (MA) could induce a deficit in neurological and neuropsychological functions [1, 2]. MA strongly affects the nervous system by enhancing the release of neurotransmitters such as dopamine (DA), serotonin, and norepinephrine [3]. MA administration demonstrates significant effects on the central nervous system (CNS), such as excessive sympathetic nervous system stimulation, neurologic complications, neuropsychiatric complications, and cognitive dysfunctions [4, 5]. Abusers can experience increased cognitive performance in acute low-dose exposure to MA [6]; however, chronic MA users come across with wide range of neuropsychological deficits [7] and cognitive impairments [4, 8-10]. MA-induced cognitive impairments mostly exhibit in the non-equally global aspect of cognition, including attention, inhibition, memory, and executive functions [11-

18]. Theoretically, MA promotes neurological and neuropsychological dysfunction via its effects of neurotoxicity and neuroinflammation.

MA can induce physiological and structural changes and altered inflammatory reactions associated with poor cognition. A study in MA abusers revealed cognitive deficits in the Stroop task accompanied by right inferior temporal gyrus hyperactivation, white matter hypertrophy, and high plasma levels of inflammatory-associated molecules releasing, including tumor necrosis factor-alpha (TNF-α) and S100 calcium-binding protein β (S100β) [19]. MA could be the critical causality of releasing molecular modulators from the CNS to the peripheral system. Additionally, the more defect in inflammatory function among MA users associates, the more exacerbate the deficit in cognition [20]. Yang and his colleagues [21] have also reported that elevated serum levels of IL-6, IL-8, and IL-10 are associated with cognitive impairments observed in MA-associated psychosis. Significant increases in serum inflammatory cytokines (TNF-α, IL-6, and IL-18) in chronic MA abusers are still observed even after abstinent from MA for almost six weeks [22].

Among ample pro-inflammatory cytokines produced by MA, IL-6 and IL-18 exclusively reported their association with cognitive impairments in various human conditions. For example, a high level of IL-6 with metabolic syndrome increases the risk of cognitive impairment [23]. Another study in post-ischemic stroke reported that patients with high IL-6 levels exhibited a high possibility of cognitive decline [24]. Notably, IL-6 negatively correlated with cognitive ability determined by Mini-Mental State Examination (MMSE) in the multiethnic cohort [25]. Additionally, the higher IL-18 is associated with more significant cognitive impairments in schizophrenia [26, 27]. The correlation between IL-18 and cognitive impairment is also found in Alzheimer's disease (AD) [28]. It seems that cytokines affect cognitive impairment in neuropsychological and neurodegenerative disorders.

Numerous studies have been taken on animals to elucidate the mechanism underlying MA-induced neuroinflammation. It has demonstrated that acute MA administration increases IL-6 and TNF-α mRNA expression in the mouse hippocampus, striatum, frontal cortex [29], and IL-1β in hippocampal tissue [30]. In addition, a study in the MA-treated rat brain demonstrates a significant increase in IL-1β mRNA expression in the hypothalamus [31]. These cytokines are related to neurotoxicity, which affirms by neurotoxic attenuation after blocking their production [30, 32, 33]. The molecular mechanism of MA elicits neuroinflammation suggested to exert the prominent role of glial cells. MA induces microglia and astrocytes activation and is related to the Toll-like receptor 4 (TLR4), which is involved in the immune surveillance of pathogens and exogenous small molecules. Consequently, TRL4 activation promotes nuclear factor-kappa B (NF-κB) to regulate the transcription of pro-inflammatory cytokines such as IL-1β, IL-6, TNF-α and IL-18 [34, 35]. These insults activate the innate and acquired immune responses and initiate inflammatory reactions in the extracellular space and contribute to the pathophysiology of neuroinflammation [36].

Under the neuroinflammatory process, pro-inflammatory cytokines can alter the tryptophan metabolite by enhancing the expression of indoleamine 2,3-dioxygenase (IDO), the key enzyme in the tryptophan metabolic pathway [37]. Then IDO converts tryptophan into kynurenic acid (KYNA), which is an N-methyl-D-aspartate receptor (NMDAR) antagonist in the human CNS [38, 39]. Elevated production of KYNA inhibits the NR1 subunit of NMDAR and alpha 7 nicotinic acetylcholine receptor (α7-nAchR), leading to decreased NMDAR function and reduced α7nAchR-mediated glutamate release [40-42]. Thus, reduced levels of glutamate and NMDAR function were observed in the prefrontal cortex [43, 44]. Furthermore, IDO enzyme activation by pro-inflammatory cytokines can also increase another tryptophan metabolite, quinolinic acid (QUIN), an NMDAR agonist. A high level of QUIN results in excitotoxicity and neural cell death [45, 46]. Thus, these tryptophan metabolites might be key elements of extensive processes under inflammatory cytokines-induced neurological and neuropsychological dysfunctions. However, the effects of MA on neuroinflammation related to tryptophan metabolites and the extent of

induced neural degeneration are not well understood.

The direct identification of target biomarkers in the human brain is limited to many ethical concerns. The interaction of neuroinflammation and tryptophan metabolites in neurodegenerative processes is mainly taken in animal studies [47, 48]. Hence, investigating biomarkers from the circulating blood system is one of the appropriately non-invasive approaches for exploring the neuropathological in the brain by taking advantage of blood-brain interaction [49]. The blood-brain barrier (BBB) could be disrupted following immune aggravation [50]; thus, biological changes discovered in the blood could trace the changes in the brain. Therefore, in this study, we aimed to determine the markers of inflammatory reactions and tryptophan metabolites in blood samples of MA abusers associated with their cognitive functions.

Methods

Participants

In this study, 15 MA abusers and 15 HC participants of the Thai population were recruited. The MA abusers were patients in Drug Abuse Treatment unit, Thanyarak hospital, Thailand. The study was approved by the Mahidol University Central Institutional Review Board (MU-CIRB 2016/043.3103). To perform experiments, all participants must previously voluntarily provide their permission in the informed consent form. The overall inclusion criteria include people 25-55 years old, normal eyesight and hearing (or had recovered to normal), and a Thai Mental State Examination (TMSE) score not less than 23. The MA addiction was diagnosed by a psychiatrist. Meanwhile, apart from cigarettes and alcohol, the HC must not represent any drug use. In addition, participants who experienced brain injury were excluded.

Questionnaires

The questionnaires contain information about demographic factors, diseases, drug administration, and drug addiction, including MA.

Screening tools

Participants were examined for psychotic symptoms by the Brief Psychiatric Rating Scale (BPRS) [51] and screening for mild cognitive impairment (MCI) by the TMSE.

TMSE is the Thai cognitive screening test developed from MMSE [52]. This Thai version screening test is purposed to screen dementia for Thai elderly and patients. The cut-off testing to distinguish dementia is 23 from 30 marks. TMSE is divided into six dimensions concerning 6 points of orientation, 3 points of registration, 5 points of attention, 3 points of calculation, 10 points of language, and 3 points of recall. Therefore, the cut-off point to determine cognitive impairment is 23 out of 30.

The BPRS is a rating scale that a clinician or researcher may use to measure 18 psychiatric symptoms, including somatic concern, anxiety, guilt feeling, grandiosity, depressive mood, hostility, suspiciousness, hallucination, unusual thought content, disorientation, conceptual disorganization, emotional withdrawal, tension, mannerism and posturing, motor retardation, uncooperativeness, blunted affect and excitement. The subjects specified each area of the symptoms in the rating range from 1-7; 1 mark represents no psychiatric sign, while 7 marks represent the most severe symptom. The total score is from 18-126 (lowest to highest). A score equal to or lesser than 36 is interpreted as a low level of psychiatric symptoms; however, a score of more than 36 demonstrates a high level of psychiatric symptoms.

Determination of cognitive functions

A computerized Stroop test: A computerized Stroop test is a tool used to assess inhibition, selective attention, and cognitive flexibility [53, 54]. The task comprises a set of word-naming colors, e.g., red, green, blue, and yellow. Those words are represented in a random colors of red, green, blue, and yellow. Thus, the task consists of 2 different modes; congruent and incongruent words. The congruence represents in the same way as color and word meaning. The incongruence illustrates different ways between color and word meaning. Participants must click the right mouse button when a congruent word appears on the screen. Conversely, participants must click the left mouse button when an incongruent word appears. This task contains 200 trials to perform within 5 min-

utes. The measurement parameters are the percentage of correct congruence, miss, incongruence, and reaction time (millisecond: msec.). From these parameters, the percentage of congruence and incongruence represent selective attention and inhibition, respectively. Cognitive flexibility is the ability to switch responses between congruence and incongruence.

Go/No-Go task: The task comprises Go and No-Go conditions for assessment attention and inhibition [55, 56]. This task consisted of a set of random numbers between 0 to 9 representing on the screen by which 0 is a target clue, and 1 is a target probe. Participants must respond Go-condition when 0 is followed by 1 by immediate clicking on the left mouse button. On the contrary, they do nothing for the No-Go condition when 0 follows with a non-target probe (2-9). The task comprises 60 events of the Go-condition, 30 events of the No-Go condition, and 20 other interfering pictures. The overall measuring time is about 5 minutes. The psychometric measurement parameters include the response percentage of Go, No-Go, miss, commission error, and reaction time (msec.). The percentage of Go correct responses represents attention.

In contrast, the miss percentage should reversely correlate with this score. The percentage of No-Go and commission error reveal response inhibition. Since commission error makes a Go response on the No-Go trial, lesser commission error represents better response inhibition.

One-back test: This test mainly uses for measuring working memory [57]. The computer screen randomly displayed one Thai letter at a time. Participants must respond to a target that represents 2 continuing orders of letters by clicking on the left mouse button. The test contains 30 events of the target (15%) from a total of 200 events. It took about 6 minutes to accomplish. The psychometric scores generated from the test are composed of the percent of correct, incorrect, and reaction time (msec.).

Wisconsin card sorting test-64 (WCST-64): The WCST-64 is designed for prefrontal brain ability determination, which mainly investigates cognitive flexibility in the face of changing schedules of reinforcement. Cognitive functions such as working memory, attention, and visual processing are involved. Besides, in case of frontal lobe damage leads to poor performance determined by this test [58-61]. The WCST-64 test contains 64 cards with different geometric designs for the participants to decide which matches the best with the stimulus cards. The criteria for matching are composed of colors (red, blue, yellow, or green), forms (triangle, star, cross, or circle), and numbers (1, 2, 3, or 4). The matching process is according to the unknown rule. In this test, the matching rule will change when the correct scores continuously reach 10 cards without warning. The test generates various psychometric scores consisting of total correct (%), total error (%), perseverative response (%), perseverative error (%), non-perseverative error (%), number of trials to complete the first category, and reaction time (msec.) [62].

Complete blood count (CBC) analysis

Whole venous blood of about 3 mL was collected in an ethylenediamine tetra-acetic acid (EDTA) tube and determined as a part of a routine laboratory test in the hospital using a Beckman Coulter AU 2700 analyzer (Brea, CA).

Blood samples measurement of inflammatory cytokines

Inflammatory markers in human serum were detected by quantitative sandwich Enzyme-Linked Immunosorbent Assay (ELISA) using paired antibodies, including IL-6 (Biolegend Inc., USA) and IL-18 (R&D Systems, USA). First, venous blood was collected and left to clot at room temperature for about 30 minutes. Then, clotted blood was centrifuged at 3,000 rpm for 15 minutes at room temperature to separate serum fractions. Next, the serum samples were kept in new tubes at -80°C until examined. Inflammatory cytokines were measured by following guide protocols. The absorbance of color reaction was read at 450 nm with a microplate reader from BioTek® Instruments (Vermont, USA) for IL-6, while IL-18 was detected via EZ Read 2000 microplate reader (Biochrom, USA). Finally, the concentration of inflammatory cytokines was calculated by comparing them with their standard curves.

Measurement of tryptophan metabolites

The serum samples were also determined of tryptophan metabolites by liquid chromatogra-

Table 1. Demographic data, TMSE and BPRS scores of HC participants and MA users

Demographic data	HC (N = 15)	MA (N = 15)
Age (years)	32.20±6.81	34.13±6.40
Gender (males/female)	8/7	6/9
Years of education	13.07±3.83	11.13±2.48
Duration of methamphetamine use (years)	-	11.80±6.11
Methamphetamine use (gram/day)	-	0.30±0.57
Abstinence duration (days)	-	12.13±1.85
TMSE	28.87±1.06	26.47±2.53**
BPRS	20.00±2.27	22.87±3.48*

Values are mean ± SD. *$P < 0.05$ and **$P < 0.01$ compared to HC.

phy with tandem mass spectrometry (LC-MS/MS). The LC-MS/MS system consists of a liquid chromatography part (Dionex Ultimate 3000) in combination with an ESI/mass spectrometer (Model Q-ToF Compact II, Bruker, Germany). All samples were kept in low-protein binding microcentrifuge tubes. In the first step, serum was overnight mixed with methanol and allowed protein aggregation before collecting the supernatant. The solvent was dried using speed vacuum centrifugation and kept at -80°C until use. Then the pellets were dissolved in H_2O containing 0.1% formic acid and centrifuged at 12,000 rpm, 4°C for 5 min. At this step, the supernatants were ready to measure. Notably, a pool tube for each group was necessary to control all discovered metabolites. In the stage of measurement, the samples run in machine DIONEX Ultimate™ 3000 Ultra High-Performance Liquid Chromatography (UHPLC) system (Thermo Fisher Scientific, USA) through Acclaim™ PolarAdvantage II C18 (PA2) column (3 µm, 120 Å 2.1 × 100 mm) Dionex Bond Silica Products (Thermo Fisher Scientific, USA) with mobile phase A (0.1% formic acid in H_2O) and B (0.1% formic acid in acetonitrile) about 20 minutes. The compounds containing either positive or negative charge were determined by mass spectrometry (Model Q-ToF Compact II, Bruker, Germany).

Statistical analysis

The normality and lognormality were applied to determine the normal distribution in every pair of comparisons for appropriate parametric and nonparametric statistical tests. In addition, the Mann Whitney and Unpaired T-test were used for the nonparametric and parametric of the two classes differentiation, respectively. These were performed using GraphPad Prism 8.0.0, GraphPad Software, San Diego, California USA, www.graphpad.com. The statistical significance is determined at *p*-values less than 0.05.

Results

Demographic data of participants

There were no differences in demographic data such as substance use information, age and years of education between the two groups of participants. However, MA abusers showed a significant decrease in TMSE scores (P = 0.0021) along with significantly higher BPRS scores (P = 0.0124) when compared to HC (**Table 1**).

Cognitive performance in healthy control (HC) participants and methamphetamine abusers (MA)

Selective attention assessment by a computerized Stroop test: The results showed a significantly lower percentage of correct congruence (P = 0.0026) in MA compared to HC. Consequently, the percentage of the miss was significantly higher (P = 0.0028) in the MA group. Moreover, MA also spent more reaction time and was significantly different compared to HC (P = 0.0236) (**Table 2**).

An inhibition assessment by Go/No-Go task: The percent of correct Go and No-Go responses were determined for assessment attention and inhibition, respectively. The results demonstrated a significantly lower percentage of No-Go correct response ($P \leq 0.0001$) and higher commission error ($P \leq 0.0001$) performed by MA users compared to HC (**Table 2**).

Working memory assessment by One-back test: The percentage of a correct response on target was determined to assess working memory. MA performed a significantly lower percentage of correct responses (P = 0.0029) than HC. The result reasonably represents along with a

Table 2. Cognitive test in HC participants and MA users

Cognitive test	HC (N = 15)	MA (N = 15)
Stroop test		
Congruence (%)	95.30±5.42	87.78±6.29**
Miss (%)	4.69±5.42	12.22±6.29**
Congruence Reaction Time (msec.)	587.60±35.53	643.10±82.32*
Go/No-Go		
Go (%)	97±4.64	93.22±7.25
Miss (%)	3.00±4.64	6.78±7.25
No-Go (%)	98.67±3.03	91.56±7.44****
Commission error (%)	1.33±3.03	8.44±7.44****
Go Reaction Time (msec.)	425.10±103.9	434.80±142.50
One-back test		
Correct (%)	78.21±15.22	57.10±19.95**
Miss (%)	21.61±15.01	42.67±19.32**
Hit Reaction Time (msec.)	612.60±142.70	647.70±191.10
WCST-64		
Total correct (%)	85.36±4.38	74.48±7.03****
Total error (%)	14.64±4.38	25.96±6.80****
Perseverative response (%)	13.47±3.36	15.17±6.14
Perseverative error (%)	9.00±1.73	11.73±4.60*
Non-perseverative error (%)	5.27±3.41	14.23±5.28****
Trials to complete the first category	11.53±1.96	18.80±9.17**
Reaction time (msec.)	2253±630.2	3461± 491.9****

Values are mean ± SD. *$P < 0.05$, **$P < 0.01$, and ****$P < 0.0001$ compared to HC.

Table 3. CBC analysis in HC participants and MA abusers

CBC parameters	HC (N = 15)	MA (N = 15)	Normal range [102]
RBCs (× 1,000,000 µl)	4.96±0.60	4.45±0.79	4.00-6.10
WBCs (× 1000 µl)	6.77±1.55	8.09±1.75*	4.40-11.30
Platelet count (× 1000 µl)	283.70±61.50	316.70±62.71	179.00-356.00
Hb (g/dL)	13.71±1.77	11.38±1.37***	12.00-16.90
Hct (%)	40.90±4.73	35.99±4.52**	37.00-51.90
Neutrophil (%)	56.60±7.88	57.98±10.05	40.00-70.30
Eosinophil (%)	3.33±2.32	5.73±5.02	0.40-9.20
Basophil (%)	0.27±0.46	0.73±0.59*	0.20-1.40
Lymphocyte (%)	35.13±6.88	33.40±5.37	20.30-48.30
Monocyte (%)	4.53±1.46	4.80±1.15	3.40-12.30
MCV (fL)	82.64±5.26	81.85±8.00	80.40-98.80
MCH (pg)	27.83±1.98	25.95±3.08	25.00-31.20
MCHC (g/dL)	33.67±0.97	31.67±0.83****	30.20-34.60

Values are mean ± SD. *$P < 0.05$, **$P < 0.01$, ***$P < 0.001$, and ****$P < 0.0001$ compared to HC. MCV: Mean Corpuscular Volume; MCH: Mean Corpuscular Hemoglobin; MCHC: Mean Corpuscular Hemoglobin Concentration; g: gram; dL: deciliter; fL: femtoliter; pg: picogram.

significantly higher percentage of miss ($P = 0.0024$) (**Table 2**).

Cognitive flexibility, concept formation, and processing speed assessment by WCST-64: WCST-64 was employed to determine executive functions, especially in cognitive flexibility. MA abusers exhibited cognitive deficits compared to HC. MA group performed a significantly lower total correct score than HC ($P \leq 0.0001$). Contradictory, MA showed a significantly higher total error (%) ($P \leq 0.0001$), perseverative error ($P = 0.0403$), non-perseverative error ($P \leq 0.0001$) and trials to complete the first category ($P = 0.0025$) than HC, respectively. In addition, a significantly longer reaction time was observed in MA compared to HC ($P \leq 0.0001$) (**Table 2**).

CBC analysis

The results of CBC analysis showed that WBC numbers significantly increased in MA groups compared to HC ($P = 0.0375$). In addition, basophil, an allergic immune cell, significantly increased in MA compared to HC ($P = 0.0227$). Apart from WBC, RBC indices also showed significant differences in the MA group compared to HC. Significant decreases in hemoglobin (Hb) ($P = 0.0004$), hematocrit (Hct) ($P = 0.007$), and mean corpuscular Hb concentration (MCHC) ($P \leq 0.0001$) were observed in MA abusers compared to HC (**Table 3**).

Serum levels of pro-inflammatory cytokines and L-tryptophan

The serum levels of IL-6 slightly increased (**Figure 1A**) while the serum levels of IL-18

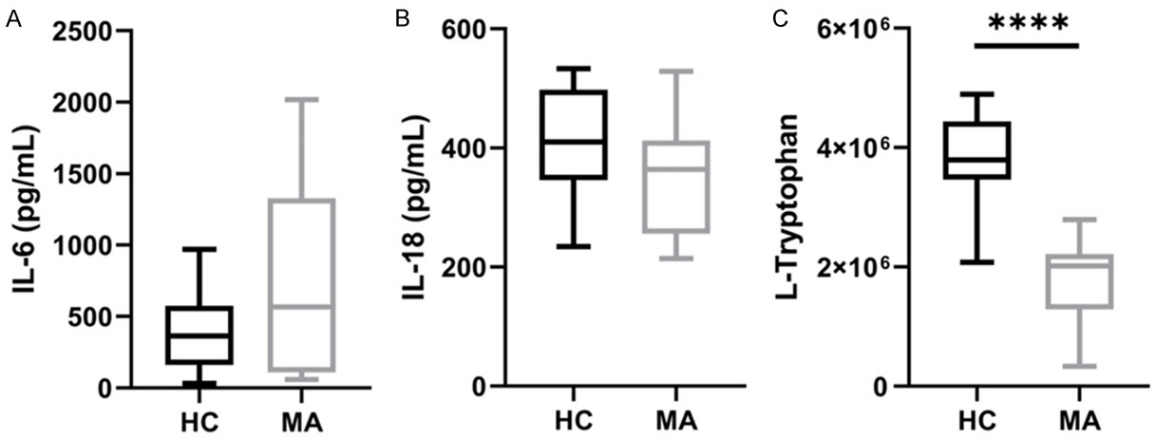

Figure 1. Serum levels of (A) IL-16, (B) IL-18, and (C) L-Tryptophan in HC participants and MA abusers. Values are mean ± SD. ****$P < 0.0001$ compared to HC.

slightly decreased (**Figure 1B**) in the MA group compared to HC, respectively. In addition, the results showed a significant decrease in L-Tryptophan levels in the MA group compared to HC ($P \leq 0.0001$) (**Figure 1C**).

Discussion

The results of the present study showed that demographic data matches HC participants and MA abusers. However, the mild cognitive assessment by TMSE [52] demonstrated low TMSE scores in MA abusers compared to the HC group. On the opposite, the evaluation of the psychotic symptoms by BPRS [51] revealed higher BPRS scores in MA abusers compared to the HC group. The neurocognitive assessment showed that MA abusers exhibited deficits in multiple cognitive functions, including selective attention, inhibition, working memory, cognitive flexibility, concept formation, and processing speed. Blood sample analysis demonstrated a decrease in RBC components, especially Hb, Hct and MCHC but an increase in WBCs and basophils in MA abusers compared to the HC group. Moreover, a slight increase in serum IL-6 levels, a slight decrease in IL-18, and a significant decrease in serum L-tryptophan levels were observed in MA abusers compared to the HC group. The key finding of this study might postulate the neuroinflammation-induced pathophysiological mechanisms of cognitive deficits in MA abusers. The inflammation in MA abusers was classified as chronic low-grade inflammation, represented by anemia of inflammation (or anemia of chronic disease) and an increase in inflammatory cytokines in blood analysis [63-66]. Taken together, the induction of inflammatory reaction was also alternatively demonstrated by the activation of tryptophan metabolites [67, 68].

Recent evidence has emphasized that oxidative damage and chronic systemic inflammation might be the initial factor to activate the anemia of inflammation. The study in phenylhydrazine (PHz)-intoxicated C57BL/6J mice model of anemia demonstrated that the antioxidant molecule, nano-complex of manganese and citrate could abolish PHz-induced reduction in Hb, RBC count, antioxidant enzyme (superoxide dismutase, glutathione peroxidase, and catalase) activities, and CD4+/CD8+ T-lymphocyte ratio, and induction in serum levels of lipid peroxidation, TNF-α, IFN-γ and IL-6 [66]. Concomitant with oxidative damage and neuroinflammation, substantial evidence has revealed that MA can induce oxidative damage and inflammation in CNS. MA in blood circulation can directly pass BBB into the brain according to its high lipid solubility [69]. The action of MA in the brain involves the generation of oxidative stress and excitotoxicity [70-72] and also induces neuroinflammation through a striking correlation to microglial activation [34, 35, 70, 73-76]. In addition, MA possesses oxidative damage by induced DA overflow and auto-oxidized, resulting in reactive oxygen species (ROS) formation [77-79]. Taken together, MA can disturb the electron transport chain in the ATP production process of mito-

chondria which also gives rise to the leakage and accumulation of ROS [70, 80]. High production of ROS further induces oxidative stress and oxidative damage. It has been reported that MA-induced oxidative stress may cause the generation of damage-associated molecular patterns (DAMPs) [81]. The correlation between MA and inflammation has been investigated for the activation of Toll-like receptors (TLRs) by DAMPs [82]. The inflammatory cascades of TLRs activation in glial cells [34, 82] subsequently induce NF-κB signaling [83] leading to an increase of inflammatory cytokines production such as IL-1β, TNF-α, IL-18, and IL-6 [29, 32, 35, 81, 82, 84-90].

Whether several lines of evidence suggest that MA can induce inflammation in CNS, leading to disturbing neurological functions; however, most studies have been done in animals [30, 91-93]. Moreover, the brain structure system is mainly closed, and manipulation of the human brain components is invasive. Thus, investigating the association between neuroinflammatory reactions and neurological function in MA abusers needs a more conclusive study. Interestingly, recent data by Tipton and colleagues suggest that MA elicits human inflammation via increased peripheral cytokine production from monocyte or macrophage [90]. Therefore, the study of blood components might be an alternative biomarker to determine the role of MA-induced neuroinflammation, which affects cognitive deficits in MA abusers.

Furthermore, our results showed a slight increase in serum IL-6 levels. Still, a slight decrease in serum IL-18 level in MA abusers compared to the HC group might explain the restorative effect of MA withdrawal [94, 95]. Thus, drug abstinence is a limitation in this study by which the more prolonged abstinence, the more recovery of brain pathology. Therefore, further study of blood inflammatory markers during MA administration are remarkably required for better elucidation.

Moreover, a new finding was observed in the present study: a significant perturbation of RBC indices such as Hb, Hct, and MCHC in MA users. These decreasing levels might be occurred by which anemia of inflammation. The evidence possibly occurs due to prolonged immune activation by several inflammatory cytokines, such as IL-1β, IL-6, IL-10, and IFN-γ which control iron homeostasis by prohibiting Hb production via promoting iron uptake into macrophage and directly damage erythroid progenitors via apoptotic activation [64]. In addition, the percentage of serum basophils up-regulated. Basophil is derived from mast cells and is responsible for allergic reactions via releasing histamine. Previous studies in rodent brains demonstrated a significant increase of histamine in the hypothalamus after a moderate dose of MA injection [96]. The same evidence might also be occurred in blood circulation as in this study. The involvement of IL-6 in the pathophysiology of anemia of inflammation has been investigated in various chronic/inflammatory diseases such as end-stage renal disease, chronic kidney disease, and rheumatoid arthritis. IL-6 mediates anemia through the induction of hypoferremia and ends with iron-restricted erythropoiesis [97, 98].

Further study on the role of neuroinflammation on neurological function has emphasized the association between low levels of tryptophan and cognitive decline [99]. The evidence of a high ratio of tryptophan metabolite, kynurenine and tryptophan was also found to conversely correlate with white matter integrity and low glutamate levels in schizophrenia [100]. Recent evidence has demonstrated that pro-inflammatory cytokines can induce the activity of IDO, the key enzyme of tryptophan metabolites, leading to decreased tryptophan levels and increased the concentration of tryptophan metabolites such as QUIN and KYNA [38, 39]. KYNA acts as an NMDAR antagonist, while QUIN acts as the NMDAR agonist in the human CNS [38, 39]. Excessive production of the tryptophan metabolic pathway results in either low glutamate levels-induced reduction in NMDAR function [40-44] or cytotoxicity-induced neural degeneration [39, 101]. This evidence might support our findings on the correlation between the low level of L-Tryptophan and cognitive deficits in MA abusers (**Figure 2**).

Taken together, the results of this study might emphasize the association between perturbations in tryptophan metabolites and low-grade systemic inflammatory conditions in MA abus-

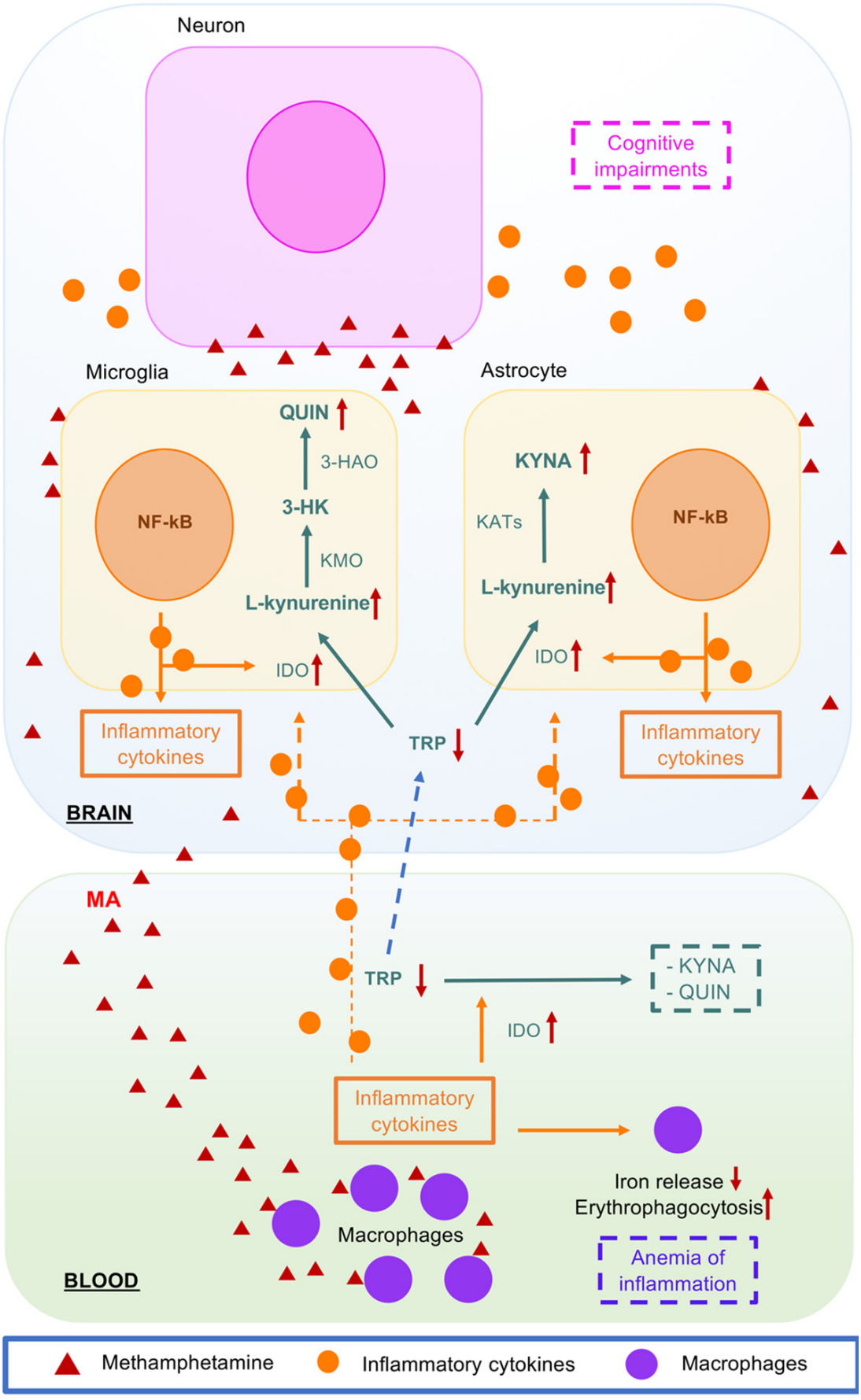
Neuron
Cognitive impairments
Microglia
Astrocyte
QUIN
3-HAO
3-HK
KMO
L-kynurenine
NF-kB
KYNA
KATs
L-kynurenine
NF-kB
IDO
IDO
Inflammatory cytokines
TRP
Inflammatory cytokines
BRAIN
MA
TRP
- KYNA
- QUIN
IDO
Inflammatory cytokines
Iron release
Erythrophagocytosis
Macrophages
Anemia of inflammation
BLOOD
Methamphetamine
Inflammatory cytokines
Macrophages

Figure 2. The postulated mechanism of blood-brain interaction in MA-induced anemia of inflammation, tryptophan metabolites and neurological dysfunction. MA affects the immune system by stimulating NF-κB cascades in macrophages in peripheral blood or microglia and astrocytes in the brain. In the brain, pro-inflammatory cytokines are produced from activated microglia and astrocytes. A similar mechanism occurs in the peripheral system by which MA stimulates pro-inflammatory cytokine production and release from macrophages. The pro-inflammatory cytokines and tryptophan might exchange between blood and the brain. Moreover, pro-inflammatory cytokines disturb iron homeostasis and Hb production in red blood cells leading to anemia of inflammation. In addition, pro-inflammatory cytokines targeting tryptophan metabolites by activating IDO1 result in the production of KYNA or QUIN. Finally, KYNA and QUIN provoke cognitive deficits by dysregulating NMDAR function, inducing cytotoxicity and neural degeneration. TRY: Tryptophan; KYNA: Kynurenic Acid; QUIN: Quinolinic Acid; 3-HK: 3-Hydroxykynurenine; KATs: Kynurenine Aminotransferase enzymes; KMO: Kynurenine-3-Monooxygenase; 3-HAO: 3-Hydroxyanthranilic Acid Oxygenase.

ers. We propose that the attribute of tryptophan metabolites in MA-induced cognitive deficits may gain further insights into their potential for developing companion diagnostics and more targeted drug interventions in neurological diseases.

Conclusion

The key findings in this study suggest that blood biomarkers of anemia of inflammation and tryptophan metabolites might be the alternative markers to demonstrate the induction of inflammatory reactions in MA abusers. Moreover, these surrogate blood inflammatory markers might utilize as an indicator to represent neuroinflammation-related neurological dysfunction in MA-induced neurotoxicity and cognitive deficits.

Acknowledgements

This study was supported by the Royal Golden Jubilee (RGJ) Ph.D. program (Grant No. PHD/0135/2560) through the National Research Council of Thailand (NRCT) and Thailand Research Fund (TRF) and Mahidol University.

Disclosure of conflict of interest

None.

Address correspondence to: Banthit Chetsawang, Research Center for Neuroscience, Institute of Molecular Biosciences, Mahidol University, Salaya, Nakhon Pathom, Thailand. Tel: 66 (0) 2441-9003-7; Fax: 66 (0) 2441-1013; E-mail: banthit.che@mahidol.ac.th

References

[1] Kraiwattanapirom N, Siripornpanich V, Suwannapu W, Unaharassamee W, Chawang O, Lomwong N, Vittayatavornwong L and Chetsawang B. The quantitative analysis of EEG during resting and cognitive states related to neurological dysfunctions and cognitive impairments in methamphetamine abusers. Neurosci Lett 2022; 789: 136870.

[2] Chen T, Su H, Zhong N, Tan H, Li X, Meng Y, Duan C, Zhang C, Bao J, Xu D, Song W, Zou J, Liu T, Zhan Q, Jiang H and Zhao M. Disrupted brain network dynamics and cognitive functions in methamphetamine use disorder: insights from EEG microstates. BMC Psychiatry 2020; 20: 334.

[3] Panenka WJ, Procyshyn RM, Lecomte T, MacEwan GW, Flynn SW, Honer WG and Barr AM. Methamphetamine use: a comprehensive review of molecular, preclinical and clinical findings. Drug Alcohol Depend 2013; 129: 167-179.

[4] Cruickshank CC and Dyer KR. A review of the clinical pharmacology of methamphetamine. Addiction 2009; 104: 1085-1099.

[5] Rusyniak DE. Neurologic manifestations of chronic methamphetamine abuse. Neurol Clin 2011; 29: 641-655.

[6] Mahoney JJ, Jackson BJ, Kalechstein AD, De La Garza R and Newton TF. Acute, low-dose methamphetamine administration improves attention/information processing speed and working memory in methamphetamine-dependent individuals displaying poorer cognitive performance at baseline. Prog Neuropsychopharmacol Biol Psychiatry 2011; 35: 459-465.

[7] Gonzales R, Ang A, Marinelli-Casey P, Glik DC, Iguchi MY and Rawson RA. Health-related quality of life trajectories of methamphetamine-dependent individuals as a function of treatment completion and continued care over a 1-year period. J Subst Abuse Treat 2009; 37: 353-361.

[8] Scott JC, Woods SP, Matt GE, Meyer RA, Heaton RK, Atkinson JH and Grant I. Neurocognitive effects of methamphetamine: a critical review and meta-analysis. Neuropsychol Rev 2007; 17: 275-297.

[9] Sabrini S, Wang GY, Lin JC, Ian JK and Curley LE. Methamphetamine use and cognitive func-

tion: a systematic review of neuroimaging research. Drug Alcohol Depend 2019; 194: 75-87.
[10] Dean AC, Groman SM, Morales AM and London ED. An evaluation of the evidence that methamphetamine abuse causes cognitive decline in humans. Neuropsychopharmacology 2013; 38: 259-274.
[11] Volkow ND, Chang L, Wang GJ, Fowler JS, Leonido-Yee M, Franceschi D, Sedler MJ, Gatley SJ, Hitzemann R, Ding YS, Logan J, Wong C and Miller EN. Association of dopamine transporter reduction with psychomotor impairment in methamphetamine abusers. Am J Psychiatry 2001; 158: 377-382.
[12] Mehrjerdi ZA, Noroozi A, Barr AM and Ekhtiari H. Attention deficits in chronic methamphetamine users as a potential target for enhancing treatment efficacy. Basic Clin Neurosci 2012; 3: 5-14.
[13] Kalechstein AD, Newton TF and Green M. Methamphetamine dependence is associated with neurocognitive impairment in the initial phases of abstinence. J Neuropsychiatry Clin Neurosci 2003; 15: 215-220.
[14] King G, Alicata D, Cloak C and Chang L. Neuropsychological deficits in adolescent methamphetamine abusers. Psychopharmacology 2010; 212: 243-249.
[15] Salo R, Nordahl TE, Galloway GP, Moore CD, Waters C and Leamon MH. Drug abstinence and cognitive control in methamphetamine-dependent individuals. J Subst Abuse Treat 2009; 37: 292-297.
[16] Salo R, Nordahl TE, Possin K, Leamon M, Gibson DR, Galloway GP, Flynn NM, Henik A, Pfefferbaum A and Sullivan EV. Preliminary evidence of reduced cognitive inhibition in methamphetamine-dependent individuals. Psychiatry Res 2002; 111: 65-74.
[17] Chung A, Lyoo IK, Kim SJ, Hwang J, Bae SC, Sung YH, Sim ME, Song IC, Kim J, Chang KH and Renshaw PF. Decreased frontal white-matter integrity in abstinent methamphetamine abusers. Int J Neuropsychopharmacol 2007; 10: 765-775.
[18] Potvin S, Pelletier J, Grot S, Hebert C, Barr AM and Lecomte T. Cognitive deficits in individuals with methamphetamine use disorder: a meta-analysis. Addict Behav 2018; 80: 154-160.
[19] Ghavidel N, Khodagholi F, Ahmadiani A, Khosrowabadi R, Asadi S and Shams J. Inflammation but not programmed cell death is activated in methamphetamine-dependent patients: Relevance to the brain function. Int J Psychophysiol 2020; 157: 42-50.
[20] Walter TJ, Iudicello J, Cookson DR, Franklin D, Tang B, Young JW, Perry W, Ellis R, Heaton RK, Grant I, Minassian A and Letendre S; On Behalf Of The Translational Methamphetamine Aids Research Center Tmarc. The relationships between HIV-1 infection, history of methamphetamine use disorder, and soluble biomarkers in blood and cerebrospinal fluid. Viruses 2021; 13: 1287.
[21] Yang X, Zhao H, Liu X, Xie Q, Zhou X, Deng Q and Wang G. The relationship between serum cytokine levels and the degree of psychosis and cognitive impairment in patients with methamphetamine-associated psychosis in Chinese patients. Front Psychiatry 2020; 11: 594766.
[22] Luo Y, He H, Ou Y, Zhou Y and Fan N. Elevated serum levels of TNF-α, IL-6, and IL-18 in chronic methamphetamine users. Hum Psychopharmacol 2022; 37: e2810.
[23] Yaffe K, Kanaya A, Lindquist K, Simonsick EM, Harris T, Shorr RI, Tylavsky FA and Newman AB. The metabolic syndrome, inflammation, and risk of cognitive decline. JAMA 2004; 292: 2237-2242.
[24] Wang Y, Li J, Pan Y, Wang M, Lin J, Meng X, Liao X and Wang Y. Interleukin-6 as predictor of one-year cognitive function after ischemic stroke or TIA. Neuropsychiatr Dis Treat 2022; 18: 391-399.
[25] Wright CB, Sacco RL, Rundek T, Delman J, Rabbani L and Elkind M. Interleukin-6 is associated with cognitive function: the Northern Manhattan study. J Stroke Cerebrovasc Dis 2006; 15: 34-38.
[26] Wu JQ, Chen DC, Tan YL, Tan SP, Xiu MH, Wang ZR, Yang FD, Soares JC and Zhang XY. Altered interleukin-18 levels are associated with cognitive impairment in chronic schizophrenia. J Psychiatr Res 2016; 76: 9-15.
[27] Zhang XY, Tang W, Xiu MH, Chen DC, Yang FD, Tan YL, Wang ZR, Zhang F, Liu J, Liu L, Chen Y, Wen N and Kosten TR. Interleukin 18 and cognitive impairment in first episode and drug naive schizophrenia versus healthy controls. Brain Behav Immun 2013; 32: 105-111.
[28] Bossù P, Ciaramella A, Salani F, Bizzoni F, Varsi E, Iulio F, Giubilei F, Gianni W, Trequattrini A, Bernardini S, Caltagirone C and Spalletta G. Interleukin-18 produced by peripheral blood cells is increased in Alzheimer's disease and correlates with cognitive impairment. Brain Behav Immun 2008; 22: 487-492.
[29] Gonçalves J, Martins T, Ferreira R, Milhazes N, Borges F, Ribeiro CF, Malva JO, Macedo TR and Silva AP. Methamphetamine-induced early increase of IL-6 and TNF-alpha mRNA expression in the mouse brain. Ann N Y Acad Sci 2008; 1139: 103-111.
[30] Liskiewicz A, Przybyla M, Park M, Liskiewicz D, Nowacka-Chmielewska M, Malecki A, Barski J, Lewin-Kowalik J and Toborek M. Methamphet-

amine-associated cognitive decline is attenuated by neutralizing IL-1 signaling. Brain Behav Immun 2019; 80: 247-254.
[31] Yamaguchi T, Kuraishi Y, Minami M, Nakai S, Hirai Y and Satoh M. Methamphetamine-induced expression of interleukin-1β mRNA in the rat hypothalamus. Neurosci Lett 1991; 128: 90-92.
[32] Ladenheim B, Krasnova IN, Deng X, Oyler JM, Polettini A, Moran TH, Huestis MA and Cadet JL. Methamphetamine-induced neurotoxicity is attenuated in transgenic mice with a null mutation for interleukin-6. Mol Pharmacol 2000; 58: 1247-1256.
[33] Yan Y, Nitta A, Koseki T, Yamada K and Nabeshima T. Dissociable role of tumor necrosis factor alpha gene deletion in methamphetamine self-administration and cue-induced relapsing behavior in mice. Psychopharmacology (Berl) 2012; 221: 427-436.
[34] Bachtell R, Hutchinson MR, Wang X, Rice KC, Maier SF and Watkins LR. Targeting the toll of drug abuse: the translational potential of toll-like receptor 4. CNS Neurol Disord Drug Targets 2015; 14: 692-699.
[35] Ojaniemi M, Glumoff V, Harju K, Liljeroos M, Vuori K and Hallman M. Phosphatidylinositol 3-kinase is involved in toll-like receptor 4-mediated cytokine expression in mouse macrophages. Eur J Immunol 2003; 33: 597-605.
[36] Cahill CM and Rogers JT. Interleukin (IL) 1beta induction of IL-6 is mediated by a novel phosphatidylinositol 3-kinase-dependent AKT/IkappaB kinase alpha pathway targeting activator protein-1. J Biol Chem 2008; 283: 25900-25912.
[37] Halaris A. Inflammation, heart disease, and depression. Curr Psychiatry Rep 2013; 15: 400.
[38] Schwarcz R and Pellicciari R. Manipulation of brain kynurenines: glial targets, neuronal effects, and clinical opportunities. J Pharmacol Exp Ther 2002; 303: 1-10.
[39] Stone TW. Neuropharmacology of quinolinic and kynurenic acids. Pharmacol Rev 1993; 45: 309-379.
[40] Hilmas C, Pereira EF, Alkondon M, Rassoulpour A, Schwarcz R and Albuquerque EX. The brain metabolite kynurenic acid inhibits alpha7 nicotinic receptor activity and increases non-alpha7 nicotinic receptor expression: physiopathological implications. J Neurosci 2001; 21: 7463-7473.
[41] Stone TW. Kynurenic acid blocks nicotinic synaptic transmission to hippocampal interneurons in young rats. Eur J Neurosci 2007; 25: 2656-2665.
[42] Marchi M, Risso F, Viola C, Cavazzani P and Raiteri M. Direct evidence that release-stimulating alpha7* nicotinic cholinergic receptors are localized on human and rat brain glutamatergic axon terminals. J Neurochem 2002; 80: 1071-1078.
[43] Wu HQ, Pereira EF, Bruno JP, Pellicciari R, Albuquerque EX and Schwarcz R. The astrocyte-derived alpha7 nicotinic receptor antagonist kynurenic acid controls extracellular glutamate levels in the prefrontal cortex. J Mol Neurosci 2010; 40: 204-210.
[44] Najjar S, Pearlman DM, Alper K, Najjar A and Devinsky O. Neuroinflammation and psychiatric illness. J Neuroinflammation 2013; 10: 43.
[45] Vecsei L, Szalardy L, Fulop F and Toldi J. Kynurenines in the CNS: recent advances and new questions. Nat Rev Drug Discov 2013; 12: 64-82.
[46] Chiarugi A, Carpenedo R, Molina MT, Mattoli L, Pellicciari R and Moroni F. Comparison of the neurochemical and behavioral effects resulting from the inhibition of kynurenine hydroxylase and/or kynureninase. J Neurochem 1995; 65: 1176-1183.
[47] Endepols H, Zlatopolskiy BD, Zischler J, Alavinejad N, Apetz N, Vus S, Drzezga A and Neumaier B. Imaging of cerebral tryptophan metabolism using 7-[^{18}F]FTrp-PET in a unilateral Parkinsonian rat model. NeuroImage 2022; 247: 118842.
[48] Orsatti L, Speziale R, Orsale MV, Caretti F, Veneziano M, Zini M, Monteagudo E, Lyons K, Beconi M, Chan K, Herbst T, Toledo-Sherman L, Munoz-Sanjuan I, Bonelli F and Dominguez C. A single-run liquid chromatography mass spectrometry method to quantify neuroactive kynurenine pathway metabolites in rat plasma. J Pharm Biomed Anal 2015; 107: 426-431.
[49] Capuron L and Miller AH. Immune system to brain signaling: neuropsychopharmacological implications. Pharmacol Ther 2011; 130: 226-238.
[50] Huang X, Hussain B and Chang J. Peripheral inflammation and blood-brain barrier disruption: effects and mechanisms. CNS Neurosci Ther 2021; 27: 36-47.
[51] Hunter EE and Murphy M. Brief psychiatric rating scale. In: Kreutzer JS, DeLuca J, Caplan B, editors. Encyclopedia of clinical neuropsychology. New York, NY: Springer New York; 2011. pp. 447-449.
[52] Train the Brain Forum Committe Poungvarin NTA. Thai mentral state examination (TMSE). Siriraj Hosp Gaz 1993; 45: 359-374.
[53] Macleod C. The stroop task: the “gold standard” of attentional measures. J Exp Psychol Gen 2002; 121.
[54] Bench CJ, Frith CD, Grasby PM, Friston KJ, Paulesu E, Frackowiak RS and Dolan RJ. Investigations of the functional anatomy of attention

using the stroop test. Neuropsychologia 1993; 31: 907-922.
[55] van Dijk F, Schellekens A, van den Broek P, Kan C, Verkes RJ and Buitelaar J. Do cognitive measures of response inhibition differentiate between attention deficit/hyperactivity disorder and borderline personality disorder? Psychiatry Res 2014; 215: 733-739.
[56] Gonthier C, Macnamara BN, Chow M, Conway AR and Braver TS. Inducing proactive control shifts in the AX-CPT. Front Psychol 2016; 7: 1822.
[57] Owen AM, McMillan KM, Laird AR and Bullmore E. N-back working memory paradigm: a meta-analysis of normative functional neuroimaging studies. Hum Brain Mapp 2005; 25: 46-59.
[58] Eling P, Derckx K and Maes R. On the historical and conceptual background of the Wisconsin card sorting test. Brain Cogn 2008; 67: 247-253.
[59] Faustino B, Oliveira J and Lopes P. Diagnostic precision of the Wisconsin card sorting test in assessing cognitive deficits in substance use disorders. Appl Neuropsychol Adult 2019; 1-8.
[60] Nyhus E and Barceló F. The Wisconsin card sorting test and the cognitive assessment of prefrontal executive functions: a critical update. Brain Cogn 2009; 71: 437-451.
[61] Sturm W. Neuropsychological assessment. J Neurol 2007; 254 Suppl 2: II12-II14.
[62] Khanthiyong B, Thanoi S, Reynolds GP and Nudmamud-Thanoi S. Association study of the functional Catechol-O-Methyltranferase (COMT) Val^{158}Met polymorphism on executive cognitive function in a Thai sample. Int J Med Sci 2019; 16: 1461-1465.
[63] Tsuboi A, Watanabe M, Kazumi T and Fukuo K. Association of low serum iron levels with low-grade inflammation and hyperadiponectinemia in community-living elderly women. J Atheroscler Thromb 2013; 20: 670-677.
[64] Weiss G, Ganz T and Goodnough LT. Anemia of inflammation. Blood 2019; 133: 40-50.
[65] Marques O, Neves J, Horvat NK, Colucci S, Guida C and Muckenthaler MU. Iron-related parameters are altered between C57BL/6N and C57BL/6J mus musculus wild-type substrains. Hemasphere 2019; 3: e304.
[66] Das M, Mondal S, Ghosh R, Biswas P, Moussa Z, Darbar S, Ahmed SA, Das AK, Bhattacharya SS, Pal D, Mallick AK, Chakrabarti P, Kundu JK, Adhikari A and Pal SK. A nano erythropoiesis stimulating agent for the treatment of anemia and associated disorders. iScience 2022; 25: 105021.
[67] Yan J, Kuzhiumparambil U, Bandodkar A, Bandodkar S, Dale RC and Fu S. Cerebrospinal fluid metabolites in tryptophan-kynurenine and nitric oxide pathways: biomarkers for acute neuroinflammation. Dev Med Child Neurol 2021; 63: 552-559.
[68] Arnhard K, Pitterl F, Sperner-Unterweger B, Fuchs D, Koal T and Oberacher H. A validated liquid chromatography-high resolution-tandem mass spectrometry method for the simultaneous quantitation of tryptophan, kynurenine, kynurenic acid, and quinolinic acid in human plasma. Electrophoresis 2018; 39: 1171-1180.
[69] Nordahl TE, Salo R and Leamon M. Neuropsychological effects of chronic methamphetamine use on neurotransmitters and cognition: a review. J Neuropsychiatry Clin Neurosci 2003; 15: 317-325.
[70] Yang X, Wang Y, Li Q, Zhong Y, Chen L, Du Y, He J, Liao L, Xiong K, Yi CX and Yan J. The main molecular mechanisms underlying methamphetamine-induced neurotoxicity and implications for pharmacological treatment. Front Mol Neurosci 2018; 11: 186.
[71] Jumnongprakhon P, Govitrapong P, Tocharus C, Tungkum W and Tocharus J. Protective effect of melatonin on methamphetamine-induced apoptosis in glioma cell line. Neurotox Res 2014; 25: 286-294.
[72] Tulloch I, Afanador L, Mexhitaj I, Ghazaryan N, Garzagongora AG and Angulo JA. A single high dose of methamphetamine induces apoptotic and necrotic striatal cell loss lasting up to 3 months in mice. Neuroscience 2011; 193: 162-169.
[73] Gonçalves J, Baptista S, Martins T, Milhazes N, Borges F, Ribeiro CF, Malva JO and Silva AP. Methamphetamine-induced neuroinflammation and neuronal dysfunction in the mice hippocampus: preventive effect of indomethacin. Eur J Neurosci 2010; 31: 315-326.
[74] Tocharus J, Chongthammakun S and Govitrapong P. Melatonin inhibits amphetamine-induced nitric oxide synthase mRNA overexpression in microglial cell lines. Neurosci Lett 2008; 439: 134-137.
[75] Tocharus J, Khonthun C, Chongthammakun S and Govitrapong P. Melatonin attenuates methamphetamine-induced overexpression of pro-inflammatory cytokines in microglial cell lines. J Pineal Res 2010; 48: 347-352.
[76] Smith JA, Das A, Ray SK and Banik NL. Role of pro-inflammatory cytokines released from microglia in neurodegenerative diseases. Brain Res Bull 2012; 87: 10-20.
[77] McDonnell-Dowling K and Kelly JP. The role of oxidative stress in methamphetamine-induced toxicity and sources of variation in the design of animal studies. Curr Neuropharmacol 2017; 15: 300-314.

[78] Halpin LE, Collins SA and Yamamoto BK. Neurotoxicity of methamphetamine and 3,4-methylenedioxymethamphetamine. Life Sciences 2014; 97: 37-44.
[79] Shin EJ, Tran HQ, Nguyen PT, Jeong JH, Nah SY, Jang CG, Nabeshima T and Kim HC. Role of mitochondria in methamphetamine-induced dopaminergic neurotoxicity: involvement in oxidative stress, neuroinflammation, and pro-apoptosis-a review. Neurochem Res 2018; 43: 66-78.
[80] Potula R, Hawkins BJ, Cenna JM, Fan S, Dykstra H, Ramirez SH, Morsey B, Brodie MR and Persidsky Y. Methamphetamine causes mitrochondrial oxidative damage in human T lymphocytes leading to functional impairment. J Immunol 2010; 185: 2867-2876.
[81] Papageorgiou M, Raza A, Fraser S, Nurgali K and Apostolopoulos V. Methamphetamine and its immune-modulating effects. Maturitas 2019; 121: 13-21.
[82] Du SH, Qiao DF, Chen CX, Chen S, Liu C, Lin Z, Wang H and Xie WB. Toll-like receptor 4 mediates methamphetamine-induced neuroinflammation through caspase-11 signaling pathway in astrocytes. Front Mol Neurosci 2017; 10: 409.
[83] Chao J, Zhang Y, Du L, Zhou R, Wu X, Shen K and Yao H. Molecular mechanisms underlying the involvement of the sigma-1 receptor in methamphetamine-mediated microglial polarization. Sci Rep 2017; 7: 11540.
[84] Clark KH, Wiley CA and Bradberry CW. Psychostimulant abuse and neuroinflammation: emerging evidence of their interconnection. Neurotox Res 2013; 23: 174-188.
[85] Shah A, Silverstein PS, Singh DP and Kumar A. Involvement of metabotropic glutamate receptor 5, AKT/PI3K Signaling and NF-κB pathway in methamphetamine-mediated increase in IL-6 and IL-8 expression in astrocytes. J Neuroinflammation 2012; 9: 52.
[86] Loftis JM, Choi D, Hoffman W and Huckans MS. Methamphetamine causes persistent immune dysregulation: a cross-species, translational report. Neurotox Res 2011; 20: 59-68.
[87] Xu E, Liu J, Liu H, Wang X and Xiong H. Role of microglia in methamphetamine-induced neurotoxicity. Int J Physiol Pathophysiol Pharmacol 2017; 9: 84-100.
[88] Lin Y, Jamison S and Lin W. Interferon-γ activates nuclear factor-κB in oligodendrocytes through a process mediated by the unfolded protein response. PLoS One 2012; 7: e36408.
[89] Wang B, Chen T, Wang J, Jia Y, Ren H, Wu F, Hu M and Chen Y. Methamphetamine modulates the production of interleukin-6 and tumor necrosis factor-alpha via the cAMP/PKA/CREB signaling pathway in lipopolysaccharide-activated microglia. Int Immunopharmacol 2018; 56: 168-178.
[90] Tipton DA, Legan ZT and Dabbous M. Methamphetamine cytotoxicity and effect on LPS-stimulated IL-1beta production by human monocytes. Toxicol In Vitro 2010; 24: 921-927.
[91] Fan R, Shen Y, Li X, Luo H, Zhang P, Liu Y, Si Z, Zhou W and Liu Y. The effect of the NLRP1 inflammasome on methamphetamine-induced cognitive impairment in rats. Drug Alcohol Depend 2022; 237: 109537.
[92] Lwin T, Yang JL, Ngampramuan S, Viwatpinyo K, Chancharoen P, Veschsanit N, Pinyomahakul J, Govitrapong P and Mukda S. Melatonin ameliorates methamphetamine-induced cognitive impairments by inhibiting neuroinflammation via suppression of the TLR4/MyD88/NFκB signaling pathway in the mouse hippocampus. Prog Neuropsychopharmacol Biol Psychiatry 2021; 111: 110109.
[93] Yang T, Zang S, Wang Y, Zhu Y, Jiang L, Chen X, Zhang X, Cheng J, Gao R, Xiao H and Wang J. Methamphetamine induced neuroinflammation in mouse brain and microglial cell line BV2: roles of the TLR4/TRIF/Peli1 signaling axis. Toxicol Lett 2020; 333: 150-158.
[94] Bossù P, Piras F, Palladino I, Iorio M, Salani F, Ciaramella A, Chiapponi C, Caltagirone C and Spalletta G. Hippocampal volume and depressive symptoms are linked to serum IL-18 in schizophrenia. Neurol Neuroimmunol Neuroinflamm 2015; 2: e111.
[95] Wu H, Zhang Z, Ma Y, Chen F, Xiong P, Xie Z, Ding G, Yu J and Wang K. Dynamic immune and exosome transcriptomic responses in patients undergoing psychostimulant methamphetamine withdrawal. Front Cell Neurosci 2022; 16: 961131.
[96] Ito C, Onodera K, Yamatodani A, Watanabe T and Sato M. The effect of methamphetamine on histamine release in the rat hypothalamus. Psychiatry Clin Neurosci 1997; 51: 79-81.
[97] Raj DS. Role of interleukin-6 in the anemia of chronic disease. Semin Arthritis Rheum 2009; 38: 382-388.
[98] Akchurin O, Patino E, Dalal V, Meza K, Bhatia D, Brovender S, Zhu YS, Cunningham-Rundles S, Perelstein E, Kumar J, Rivella S and Choi ME. Interleukin-6 contributes to the development of anemia in juvenile CKD. Kidney Int Rep 2018; 4: 470-483.
[99] Ramos-Chávez LA, Roldán-Roldán G, García-Juárez B, González-Esquivel D, Pérez de la Cruz G, Pineda B, Ramírez-Ortega D, García Muñoz I, Jiménez Herrera B, Ríos C, Gómez-Manzo S, Marcial-Quino J, Sánchez Chapul L, Carrillo Mora P and Pérez de la Cruz V. Low serum tryptophan levels as an indicator of global cognitive performance in nondemented wom-

en over 50 years of age. Oxid Med Cell Longev 2018; 2018: 8604718.
[100] Chiappelli J, Postolache TT, Kochunov P, Rowland LM, Wijtenburg SA, Shukla DK, Tagamets M, Du X, Savransky A, Lowry CA, Can A, Fuchs D and Hong LE. Tryptophan metabolism and white matter integrity in schizophrenia. Neuropsychopharmacology 2016; 41: 2587-2595.
[101] Frumento G, Rotondo R, Tonetti M, Damonte G, Benatti U and Ferrara GB. Tryptophan-derived catabolites are responsible for inhibition of T and natural killer cell proliferation induced by indoleamine 2,3-dioxygenase. J Exp Med 2002; 196: 459-468.
[102] Wongkrajang P, Chinswangwatanakul W, Mokkhamakkun C, Chuangsuwanich N, Wesarachkitti B, Thaowto B, Laiwejpithaya S and Komkhum O. Establishment of new complete blood count reference values for healthy Thai adults. Int J Lab Hematol 2018; 40: 478-483.