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Recent updates on drug abuse analyzed by neuroproteomics studies: Cocaine, Methamphetamine and MDMA

Firas Kobeissy[a,1,*], Tarek H. Mouhieddine[b,1,*], Amaly Nokkari[c], Muhieddine Itani[d], Mohammed Mouhieddine[e], Zhiqun Zhang[a], Rui Zhu[f], Mark S. Gold[a], Kevin K. Wang[a,*], Yehia Mechref[f,*]

[a] Department of Psychiatry, Center for Neuroproteomics & Biomarker Research, Departments of Psychiatry and Neuroscience, University of Florida, Gainesville, FL, USA
[b] Faculty of Medicine, American University of Beirut Medical Center, Beirut, Lebanon
[c] Department of Biochemistry and Molecular Genetics, Faculty of Medicine, American University of Beirut, Beirut, Lebanon
[d] Faculty of Medicine, Saint George University of London, Nicosia, Cyprus
[e] Department of Chemical Engineering, American University of Beirut, Beirut, Lebanon
[f] Department of Chemistry and Biochemistry, Texas Tech University, Lubbock, TX, USA

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ABSTRACT

Currently, drug abuse and addiction represent a global public health concern with about 13.6 million people using illicit drugs in the USA alone. Substance abuse intervenes in normal brain functioning, causing alterations in memory, behavior and neuronal physiology. Although many studies have been conducted to elucidate the mode of action of different drugs, the heterogeneous modes of drug intake led to a complicated profile of drug-induced brain changes involving neurotoxicity and addiction. Given the complex interplay of genes and proteins in mediating these effects, neuroproteomics analysis has been considered among the methods of choice to complement what has already been discovered and to create targeted therapies. In this review, we will focus on three drugs, namely cocaine, methamphetamine (METH) and 3,4-methylenedioxy-N-methylamphetamine (MDMA). In the context of neuroproteomics, these drugs have been extensively studied by utilizing different experimental models, including primate and non-primate animals along with postmortem human samples. Even though there are many variations in the results, these drugs were shown

Abbreviations: MDMA, 3,4-methylenedioxy-N-methylamphetamine; METH, methamphetamine; MDA, 3,4-methylenedioxyamphetamine; HHA, 3,4-dihydroxyamphetamine; HHMA, 3,4-dihydroxymethamphetamine; MS, mass spectrometry; 2D-DIGE, 2-dimensional differential gel electrophoresis; HPLC, high performance liquid chromatography; LC, liquid chromatography; 2D-PAGE, 2D polyacrylamide gel electrophoresis; SDS-PAGE, sodium dodecyl sulfate-PAGE; MALDI TOF, matrix assisted laser desorption/ionization-time of flight; SELDI, surface enhanced laser desorption/ionization; ESI, electrospray ionization; iTRAQ, isobaric tags for relative and absolute quantitation; CAX, cation–anion exchange chromatography; RPLC, reversed phase liquid chromatography; MRM, multiple reaction monitoring; SILAC, stable isotope labeling with amino acids in cell culture; PMF, peptide mass fingerprinting; CPP test, conditioned place preference test; ip, intra-peritoneal; IV, intra-venous; sc, subcutaneous; NAc, nucleus accumbensm; PFC, medial prefrontal cortex; OFC, orbitofrontal cortex; DA, dopamine; DAT, dopamine transporter; GABA, γ-aminobutyric acid; COD, cocaine overdose; PC12, rat pheochromocytoma cells; cAMP, cyclic AMP; COC, cocaine; SAL, saline; NADH, nicotinamide adenine dinucleotide; NDFUS2, NADH dehydrogenase [ubiquinone] flavoprotein 2; NDFUS11, NADH dehydrogenase [ubiquinone] flavoprotein 11; GSTO1, glutathione transferase omega-1; NDUFA10, NADH dehydrogenase [ubiquinone] 1-alpha subcomplex subunit 10; DRP2, dihydropyrimidinase-related protein 2; PRDX2, peroxiredoxin-2; PRDX5, peroxiredoxin-5; PEBP1, phosphatidylethanolamine-binding protein 1; FAH2A, fumarylacetoacetate hydrolase domain containing 2A; ERK3, extracellular signal-regulated kinase 3; DDAH1, dimethylarginine dimethylaminohydrolase 1; CHP, calcium-binding protein P22-like; CNRIP1, CB1 cannabinoid receptor-interacting protein; AKAP, A-kinase anchoring protein; AMPA, α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid; NMDAD, -aspartate; GFAP, glial fibrillary acidic protein; GRP78/BiP, 78 kDa glucose-regulated protein/binding immunoglobulin protein; C/Ebp, cytidine-cytidine-adenosine-adenosine-thymidine/enhancer-binding proteins; Bcl-2, B-cell lymphoma 2; Bcl-xL, B-cell lymphoma-extra large; Bax, Bcl-2 associated X protein; Bad, Bcl-2-associated death promoter; Bid, BH3 interacting domain death agonis; tDLP1, dynamin-like protein 1; UCH-L1, ubiquitin carboxy-terminal hydrolase L1; GDI, GDP dissociation inhibitor; PEBP, phosphatidyl ethanolamine-binding protein; CAP1, adenylyl cyclase-associated protein 1; SNAP-25, synaptosomal-associated protein 25; TP1, transition protein 1; LDH-B, lactate dehydrogenase B; Tpi1, triosephosphate isomerase 1; GSTP, Gapd, glutathione S-transferase pi; SOD1, mitochondrial Cu−Zn superoxide dismutase; DDAH1, dimethylarginine dimethylaminohydrolase 1; ADMAL, -arginine; NOS, nitric oxide synthase; MAP-τ, microtubule associated protein tau; SBDP, αII-spectrin breakdown product; LC3, light chain 3; MAPK, mitogen activated protein kinase; MKK1, MAP kinase kinase 1; CRMP2, collapsin response mediator protein 2; PKC, activating protein kinase C; MRM, multiple reaction monitoring; SPTN1, spectrin alpha chain non-erythrocytic 1; SV2A, synaptic vesicle glycoprotein 2A; MYPR, myelin proteolipid protein; BASP1, brain acid soluble protein 1; VATB2, V-type proton ATPase subunit B brain isoform; GSTA3, glutathione S-transferase alpha 3; FETUA, alpha-2-HS-glycoprotein; K6PP, 6-phosphofructokinase type C; SCRN1, secernin-1; GNAI, guanine nucleotide-binding protein G(i) subunit alpha; HSP, heat shock protein; CYP, cytochrome P450; l-NNAL, -arginine; MPTP, 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine; DSP-4, N-(2-chloroethyl)-N-ethyl-2-bromobenzylamine; VMAT-2, vesicular monoamine transporter 2 protein; SERT, serotonin transporter.

[1] Firas Kobeissy and Tarek H. Mouhieddine have contributed equally to this work.

* Corresponding author. Kevin K.W. Wang, PhD and Firas Kobeissy, PhD University of Florida, Department of Psychiatry, 100 S Newell drive Rm: L4-100, Gainesville, Fl, 32611, USA
* Corresponding author. Tarek H. Mouhieddine, MD candidate Faculty of Medicine, American University of Beirut Medical Center, Beirut, Lebanon Tel: +96170630474;
* Corresponding author. Yehia Mechref, PhD Department of Chemistry and Biochemistry, Texas Tech University, Lubbock, TX 79409-1061, 806-834-8246806 742 1289.
E-mail addresses: firasko@gmail.com (F. Kobeissy) tarek.mouhieddine@gmail.com (T.H. Mouhieddine) kwang@ufl.edu (K.K. Wang) yehia.mechref@ttu.edu (Y. Mechref).

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to employ common pathways in eliciting their effects. Neuroproteomics analysis of these drugs has led to the identification of differentially expressed proteins involved in metabolism, oxidative stress, cell signaling, cytoskeleton, cell death and synaptic plasticity. Finally, this work will discuss recent findings from our laboratory by looking at a model of chronic methamphetamine abuse and its effect on different brain regions.

1. Introduction

1.1. Drug abuse and addiction

Nowadays, drug abuse and addiction are among the prominent public health concerns, with a staggering number of 23.9 million people using illicit drugs and 4.5 million dependent on them in the USA alone [160]. Furthermore, there are only a few pharmacologic therapies that are specific for drug abuse and addiction [161]. Substances of addiction highly influence brain neurotransmissions and amplify the severity of medical conditions, such as diabetes and cardiovascular diseases [1]. Drug abuse, for instance, intervenes with normal brain networking and causes severe alterations in behavior, memory and neural cell life [2] and may even cause neuronal cell death [3]. This is why it is essential to understand the complex mechanisms through which every drug inaugurates its effect on a broad spectrum of cell signaling pathways and proteins, and this is where proteomics come into play. Addiction is defined as a brain pathology provoked by complex processes that include physiological changes in certain brain regions such as the hippocampus, ventral tegmental area, nucleus accumbens (NAc), and prefrontal cortex [4]. Interestingly, the effects of drug abuse were shown to be similar to those of traumatic brain injury (TBI). Lange et al. assessed the cognitive measures of verbal and visual memory and executive functioning abilities among 104 patients with mild TBI compared to 104 drug-abusers, and it was determined that there were no apparent differences among those two groups [5]. In this paper, we discuss recent updates in the fields of neuroproteomics in drug abuse followed by a brief discussion of our own lab findings of a model of chronic methamphetamine (METH) abuse and its effect on different brain regions. The search criteria for the literature review included a PubMed search for the term "Neuroproteomics" along with the different drug names. Only the most recent neuroproteomics studies were taken into account and discussed in this paper.

1.2. Proteomics

Proteomics is an analytical approach enabling the analysis of proteins encoded by the genome of an organism [6]. A unit gene can generate several proteins via DNA processes such as transcription and translation. Proteomic analysis was introduced because the correlation between mRNA and protein levels proved insufficient in studying protein expression levels from mRNA data [7]. Proteomic approaches can be further divided into four categories: 'interaction proteomics', 'expression proteomics', 'structural proteomics' and 'functional proteomics' [8].

Proteomic studies fundamentally enable the identification and quantification of hundreds of proteins, thereby facilitating the global analysis of the function, variation, configuration and dynamics (localization, interaction and post-translational modifications) of proteins [9]. Thus, a proteomic study is ideal to characterize the cellular and molecular mechanisms of neuropsychiatric maladies, including drug abuse and addiction [2]. Herein, proteomic analysis reveals the pathophysiological routes involved [10] and unravels the behavioral alterations associated with these disorders [11]. Furthermore, drug response in addicts can be predicted and a therapeutic scheme can be prescribed and monitored according to the observed changes in the protein expression in humans and animal models, through proteomic analysis [12].

Proteomes include over 13 million distinct proteins in all species and over two million distinct proteins in humans alone [9]. Clinical applications of proteomics involves assessing the proteins in tissues, cells and body fluids, including "human plasma, serum, urine, cerebrospinal fluid, nipple aspirate fluid, ductal lavage, amniotic fluid, bile, lymph, breast milk, mucus, pleural fluid, saliva and tears" [13]. Rapid advancements in proteomic research primarily hinges on the enhanced sensitivity of mass spectrometry (MS), bioinformatics, systems biology and genomic sequence data acquisition [14].

1.3. Proteomic limitations in neuropsychiatry

A proteomic study is currently challenged by a number of analytical limitations, making proteomic-based studies far more complex than genomic-based ones [15]. One limitation is that in brain tissues, as in other types of tissues and cell lines, it is quite difficult to associate mRNA expression with protein translation [16,17]. In addition, few high-concentration proteins dominate each type of tissues, making the analysis of the low-abundant proteins challenging [18]. Moreover, thousands of copies of interrelated splices from a unit gene are found in the brain [19]. Another limitation of proteomics is that neuropsychiatric disorders are multigenic by nature and include the interaction of multiple proteins that are expressed spatially on a small scale and are in different stages of the psychological disorder [20]. The solubility of surface hydrophobic proteins, limited separation range of proteins and limited detection sensitivity of analytical techniques also pose a challenge in the proteomics field [21]. Herein, unlike genomic studies that can study tens of thousands of gene expressions at a time, proteomic studies can only study a few thousands at a time [22]. The main challenge in animal studies pertains to sample collection and preparation, where sample size is negligible and thus may not be suitable for proteomics analysis. This problem is translated in the brain regions such as the hippocampus and the thalamus which are limited in size, hard to obtain in significant amounts and, in addition to all other brain regions, are susceptible to degradation by various cellular proteases and sample collection variations [23].

1.4. Proteomic tools and methodology

The basic steps of any proteomic method include extracting proteins from tissues, applying a separation/fractionation technique to transform complex protein mixtures into ones of fewer components, analyzing the sample via an advanced technique such as MS and processing the obtained data by comparing the results with those found in the databases or literature [24,25]. A schematic of the general methodology followed by the proteomic studies discussed in this paper is depicted in Fig. 1.

To address the issue of protein abundance as a limiting step in the proteomic approach, fractionation steps are essential before starting

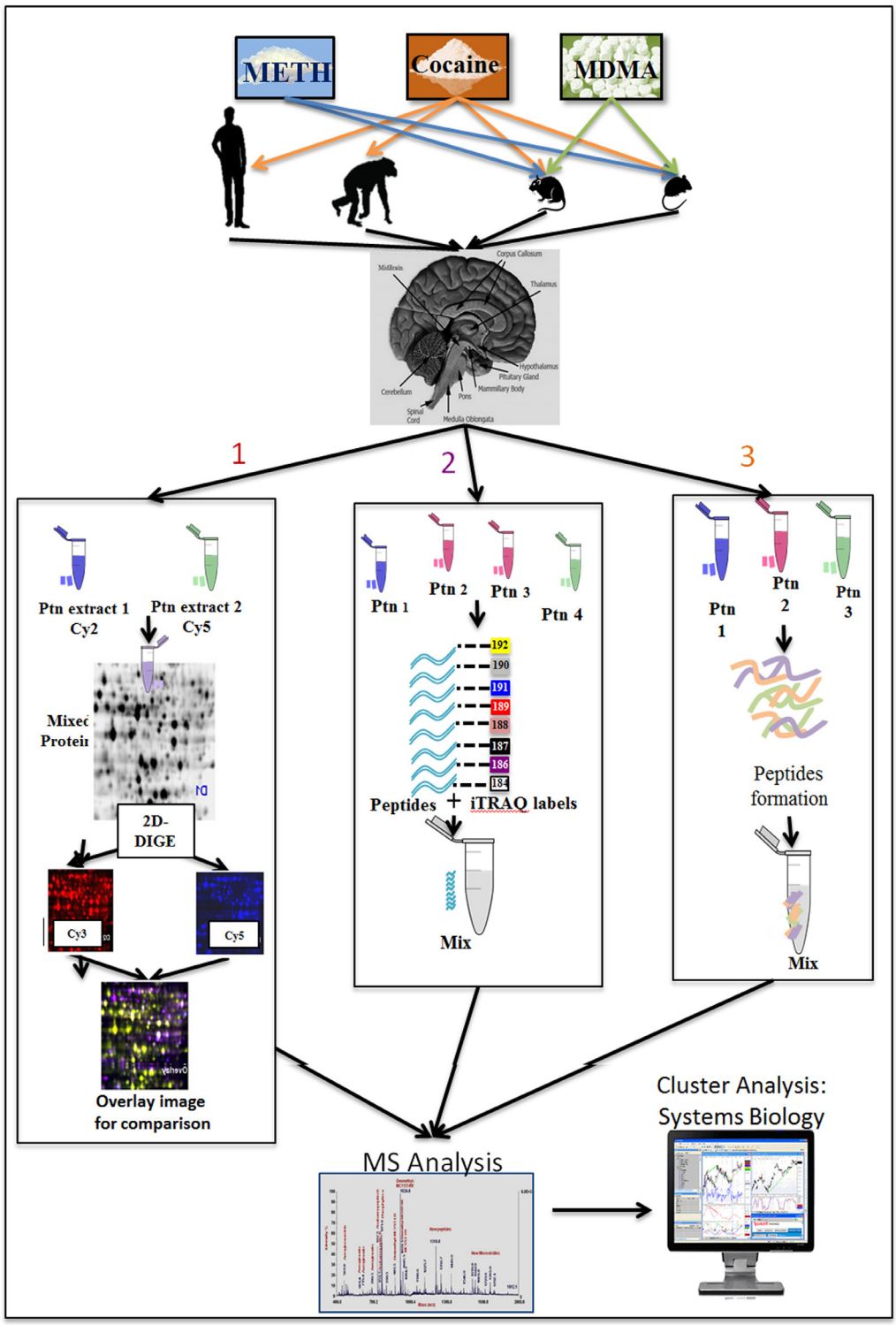

Fig. 1. Schematic of the general procedures followed by the proteomic studies discussed in this paper (1) 2D-DIGE technique, (2) iTRAQ technique and (3) Shotgun analysis.

the MS analysis [26]. Even highly abundant serum proteins may be exhausted using affinity chromatography-based techniques such as 2D polyacrylamide gel electrophoresis (2D-PAGE) and high performance liquid chromatography (HPLC) [27].

MS is an important method in proteomic analysis. It can be commenced by either electrospray ionization (ESI) or matrix-assisted laser desorption/ionization (MALDI) methods, which are both "soft ionization" techniques that transfer proteins into the gaseous state without too much fragmentation [14,28]. For simple peptide samples, examination with MALDI-time of flight (TOF) MS is acceptable, whereas, for more complex systems, separation using HPLC column joined to ESI MS systems is recommended [29]. Surface enhanced laser desorption/ionization (SELDI) MS is based on retaining proteins through protein chip systems with pre-activated surfaces via antibodies, receptors, ionic or hydrophobic materials followed by ionization and MS detection [30].

Another quantitative method is iTRAQ, also known as isobaric labeling for relative and absolute quantitation. iTRAQ is a procedure in which samples are tagged disjointedly with isobaric labels before being blended and introduced to liquid chromatography and tandem MS (LC/MS/MS) [31]. The isobaric labels comprise an abundance reporter ion, which is directly related to the protein quantity they are associated with [32].

Another approach is to use shotgun proteomics as an analytical method, which allows for the identification and quantitation of hundreds of complex sample proteins [33]. Proteins are first digested by enzymes such as trypsin and are then sent to MS system for analysis [34]. Some advantages of this method include: basic protein analysis, high molecular weight protein analysis (>120 kDa) and low abundance protein detection [35].

Finally, two-dimensional difference gel electrophoresis (2D-DIGE) is among the basic traditional multidimensional methods, which enables the visual assessment of proteins. In 2D-DIGE, proteins extracted from two or three samples are tagged directly with different fluorescent dyes such as Cy2, Cy3 and Cy5 before being separated by 2-DE [36]. The dynamic changes in protein levels related to various states can be easily detected, thus generating sensitive and reproducible results [37]. Some disadvantages associated with this method are the solubility of the hydrophobic protein surface, the restricted dynamic range and the use of amine-reactive DIGE dyes for labeling [21].

Post MS analysis and data acquisition, spectral library tools are used to compare MS scans to those stored in these libraries. In the medical field, there is still a shortage of information in protein and peptide databases for identification, functional and structural proteomic analysis [38]. Thus, there is a need to continue developing competent tools for proteomic analysis.

Below is a review of recent studies in the field of neuroproteomics pertaining to the effect of drug abuse and addiction on various brain regions. Table 1 summarizes the major neuroproteomic studies discussed.

2. Cocaine neuroproteomics

2.1. General overview

Cocaine is globally known as one of the most dangerous illicit drugs with high addictive properties, and it is believed that over 1.6 million Americans are currently using cocaine, including males and females aged 12 years or older (SAMSHA, 2013). Cocaine misuse is an intricate behavior that is influenced by genetic and environmental factors, which in turn reflects different levels of cocaine abuse vulnerability among humans and animal models. It is a chronic mental disorder in which the addict compulsively consumes drugs irrespective of the consequences, leading to the progression of drug seeking behavior [40]. Even though drug addiction studies have indicated that cocaine's oxidative metabolites are among the causes behind its negative side effects [59], the biochemical pathways behind urging an individual to seek and abuse cocaine, and other drugs, are still to be fully elucidated.

Importantly, many studies have demonstrated that the NAc invigorates the euphoric effects of cocaine [39,42,43]. More specifically, several studies have indicated that cocaine up-regulates the cyclic AMP (cAMP) pathway [60], glutamate [61], activator protein 1 family members [62], dopamine (DA) [63], γ-aminobutyric acid (GABA), growth factors, opiate receptors, metabolism and cytoskeletal elements present in the NAc [64–67].

To assess the effects of the different psychoactive drugs in animal models, researchers employ a number of behavioral testing among which is the conditioned place preference (CPP) test. In this model, an animal is placed in a specific environment that is paired with the administration of a certain drug, followed by an assessment of the animal's preference towards that environment. Thus, an animal spending an increased time in the drug-associated environment correlates with the positive rewarding properties of the drug under study [68]. In other words, this method can simulate drug addiction with its different stages from attainment to extinction and reinstatement.

2.2. Neuroproteomics in rodent models

In one study by Del Castillo et al., rats underwent cocaine-induced CPP (15 mg/kg intraperitoneally (ip)) and were separated into two subpopulations according to whether drug addiction was extinguished (E) or not extinguished (NE) [39]. Following an extra administration of either cocaine (COC) or saline (SAL), 2D-DIGE and MALDI-TOF MS were used to profile the proteome of the rats' NAc. When comparing E SAL rats to NE SAL rats, NE SAL rats had a decreased expression of the three metabolic proteins (ATP synthase alpha subunit, transketolase and fumarate hydratase) and an increased expression of NADH dehydrogenase [ubiquinone] flavoprotein 2 (NDFUS2) and glutathione S-transferase omega-1 (GSTO1). In contrast to this study, Guindalini et al. found that there is an increased glutathione transferase activity due to a genetic polymorphism detected more frequently in cocaine users compared to healthy individuals [59]. Thereafter, when comparing E SAL to E COC, Del Castillo et al. identified three out of six proteins: an up-regulated NDUFA10 and a down-regulated fumarate hydratase and dihydropyrimidinase-related protein 2 (DRP2), where DRP2 has been previously discovered to aid in axon elongation when phosphorylated [69]. Remarkably, the down-regulation of fumarate hydratase in E COC and NE SAL, when compared to E SAL, suggested that a single dose of cocaine might reinstate the cocaine-seeking behavior in rats via a mechanism pertaining to fumarate hydratase. Also, when comparing NE COC to E COC, the proteins ATP5a1, NDUFA10, peroxiredoxin-5 (PRDX5), and nucleoside diphosphate kinase B were decreased while the 60 kDa heat shock protein (HSP60) was increased. Finally, comparing NE COC to NE SAL also yielded a difference in protein expression. The identified proteins were peroxiredoxin-2 (PRDX2), (PRDX5), GSTO1, NDUFS2, α-synuclein, phosphatidylethanolamine-binding protein 1 (PEBP1) and fumarylacetoacetate hydrolase domain containing 2A (FAH2A). Of interest is a previous study performed by Mash et al., which established that α-synuclein blood level reduction via cocaine diminishes drug craving intensity [70]. Moreover, in a recent study by Ryan et al. it was demonstrated that α-synuclein can also be involved in decreasing dopaminergic transmission [71]. Finally, Goumon et al. found that PEBP1, a Raf kinase inhibitor, is expressed in the limbic system [72].

In another study conducted by Guan and Guan, rat medial prefrontal cortex (mPFC) was evaluated following repeated cocaine exposure (10 mg/kg ip) [40]. Proteins were separated via 2DE, identified and quantified via MALDI-TOF MS and tandem TOF/TOF MS. Two groups of rats (either being injected with cocaine or saline) underwent CPP for 8 days to discard rats that spend more than 500 s in the same chamber, followed by a CPP test on the ninth day to evaluate cocaine-induced reward effects. Using 2DE, they found that 50 spots were significantly up-regulated and 21 were significantly down-regulated in the group of cocaine treated rats. Moreover, Guan and Guan confirmed the presence of that same trend of some differentially expressed proteins via western blotting, which added credibility to their work. Even though neuroproteomics is a very sensitive and accurate method, it would still be more favorable to confirm the obtained results by performing traditional techniques such as western blot, ELISA and PCR. Furthermore, in addition to the metabolic proteins, more than 27% of the total mapped proteins of this study were related to actin cytoskeleton regulation where there was an increase in stathmin and tubulin alpha 8 chains but a decrease in the tubulin alpha 3 chain. Thus, the findings of this study show that cocaine addiction may lead to structural and functional impairments in the mPFC by disrupting axonal transport where Duncan et al. have

Table 1
List of recent neuroproteomics studies on the effect of cocaine, METH and MDMA on different brain regions.

Drug	Model	Drug dosage	Brain region	Proteomic method	Number of identified differentially expressed proteins	Functional domains of proteins	Reference
Cocaine	Rats	15 mg/kg for 3 days	NAc	2-DE and MALDI-TOF MS	14	Mitochondrial, dopaminergic transmission, anti-oxidant, cell structure and cell signaling	Del Castillo et al. [39]
	Rats	10 mg/kg of chronic administration	mPFC	2-DE and MS	71	Metabolism, cell structure, anti-oxidants, cell signaling, synaptic proteins, detoxification, structure regulatory proteins, cell cycle regulatory proteins, proteinase, ligase, transferase, gene regulatory protein, sensor protein and others	Guan and Guan [40]
	Rats	10 days of 40 infusions/day of 1.5 mg/kg per infusion	mPFC	2D-DIGE and MALDI-TOF/TOF MS	20	Cell structure and metabolism	Lull et al. [22]
	Rats	2 weeks of self administration of 0.05 ml of 0.2 mg cocaine with each lever press	NAc	iTRAQ	42	Cytoskeleton, synaptic, cell signaling, channels, receptors, transporters, metabolism and cell adhesion	Reissner et al. [41]
	Juvenile Vervet Monkeys	14 days of chronic administration of 2 or 4 mg/kg/day	OFC	2D-DIGE and iTRAQ	40	Cytoskeleton, metabolism, cell cycle, synaptic, protein folding, cell adhesion and cell signaling	Olausson et al. [31]
	Rhesus Monkeys	Chronic administration of 0.5 mg/kg	NAc	2D-DIGE and MALDI-TOF/TOF MS	18	Cell structure, metabolism and cell signaling	Tannu et al. [42]
	Post-mortem human samples of cocaine overdose victims	N/A	NAc	2D-DIGE and MALDI-TOF MS	15	Cell structure, synaptic plasticity, signal transduction, mitochondrial and metabolic	Tannu et al. [43]
Methamphetamine	Rats	Acute administration of 1 mg/kg	Striatum	2DE and MALDI-TOF MS	26	Mitochondrial, metabolic, protein degradation and cell structure	Iwazaki et al. [44]
	Rats	Saline injection first 2 days, followed by METH (1 mg/kg) on 3rd day and then 4 mg/kg for 10 days	Striatum	2-DE and MALDI-TOF MS	32	Metabolism, cytoskeleton, oxidative, mitochondrial, synaptic and apoptosis	Iwazaki et al. [45]
	Rats	Acute administration of 1 mg/kg	Amygdala	2DE and MALDI-TOF MS	48	Metabolism, cellular cytoskeleton, oxidative stress and apoptosis	Iwazaki et al. [46]
	Rats	8 injections of 15 mg/kg	Hippocampus, striatum and frontal cortex	2DE and MALDI-TOF/TOF MS	30	Oxidative stress, protein degradation, scavengers, mitochondrial, metabolism and apoptosis	Li et al. [47]
	Rats and PC12	Acute administration of 15 mg/kg	Striatum	2DE and MALDI-TOF/TOF MS and SILAC	30 Nitrated proteins	Cytoskeleton, cdk5 activation, ribosome function, protein nitration and apoptosis	Zhang et al. [48]
	Rats	Acute administration of 10, 20 or 40 mg/kg	Cortex and hippocampus	SDS-PAGE	AII-spectrin and tau	Cytoskeleton	Warren et al . [49]
	Rats	Acute administration of 40 mg/kg	Cortex	CAX-1D-SDS-PAGE and RPLC MS/MS	82	Oxidative stress, synaptic transmission and cell death	Kobeissy et al. [50]
	Rats	3 discontinuous injections of 1 mg/kg	NAc, PFC, cingulate cortex, striatum and hippocampus	2-DE and MALDI-TOF MS	27	Cytoskeleton, cell signaling, transport, endocytosis and exocytosis	Yang et al. [51]
	Rat	8 injections of 1 mg/kg	Frontal cortex	2-DE and MS/MS	47 Cytosolic and 42 membrane-bound proteins	Metabolism, redox regulation, protein degradation, cellular growth, synaptic function and cytoskeleton	Faure et al. [52]
	Mice	3 injections of 10 mg/kg	Striatum and susbtantia nigra	2-DE and Reverse phase-nano-HPLC-ESI-MS/MS	5	Mitochondrial and protein degradation	Liao et al. [53]

Table 1
Continued

Drug	Model	Drug dosage	Brain region	Proteomic method	Number of identified differentially expressed proteins	Functional domains of proteins	Reference
MDMA	Rats	5 mg/kg for 10 days	Hippocampus	SDS-PAGE and MALDI-TOF MS	8	Intracellular signaling, neuroplasticity, transport and cytoskeleton	van Nieuwenhuijzen [54]
	Rats	Administration of 10, 20 and 30 mg/kg	Striatum	N/A	N/A	Mitochondrial	Puerta et al. [55]
	Rats	100 mg/kg of DSP-4 and 4 administrations of 10 mg/kg MDMA	Hippocampus	SDS-PAGE	N/A	SERT and VMAT-2	Biezonski and Meyer [56]
	Rats	2 oral administration of 10 mg/kg	Liver	2-DE and Reverse phase-nano-LC-tandem MS	33	Oxidation and nitration	Moon et al. [57]
	Rats	2 oral administration of 10 mg/kg	Liver	2-DE and Reverse phase-nano-LC-tandem MS	22	Cell signaling, apoptosis, anti-oxidant, metabolism, mitochondrial and calcium regulation	Upreti et al. [58]

shown that stathmin is important in maintaining microtubule integrity [73]. Other up-regulated proteins include the ERK3 protein, a protein that has proved to be disruptive of signal transduction and neuronal morphology [74] and dimethylarginine dimethylaminohydrolase 1 (DDAH1), which was shown to affect nitric oxide signal transduction and plays a role in cocaine CPP memory reconsolidation [75].

It is noteworthy that calcium-binding protein P22-like (CHP) was increased, which means that calcineurin may have been inhibited. This inhibition was previously shown to modulate synapsin 1 protein [76] that is located on the presynaptic nerve terminals and involved in DA release post psychostimulant administration [77]. Other up-regulated proteins related to synaptic plasticity and trafficking, identified by Guan and Guan, include neuroligin 4, neurexin beta 1, synaptotagmin 15, butyrophilin 1 and CB1 cannabinoid receptor-interacting protein (CNRIP1). All of these results suggest that cocaine may lead to neuro-adaptations of drug-associated synaptic plasticity, morphological and structural changes in the mPFC and can in turn contribute to the start and augmentation of drug seeking and abuse [78].

Taken together, it can be noticed that even though Del Castillo et al. and Guan and Guan identified similar proteins in both the NAc and mPFC, respectively, each brain region differentially expressed a specific subunit of each of those proteins. For example, cocaine induced a differential expression of ATP synthase beta subunit, NDFUS11 and the ATP synthase lipid binding protein in the mPFC. In contrast, the same drug induced a differential expression of the ATP synthase alpha subunit and NDFUS2 in the NAc.

In another study by Reissner et al., NAc tissues were analyzed in a rat model of cocaine administration (intra-jugular 40 infusions/day of 1.5 mg/kg per infusion for 10 days) using iTRAQ proteomic platform, thus identifying 42 proteins of the postsynaptic density-enriched subfraction that are differentially expressed [41]. Focus was particularly given to the up-regulated AKAP79/150 protein. This protein was characterized by Wong and Scott, as a postsynaptic density scaffolding protein that confines signaling molecules to the synapse [79]. Overall, the findings of Reissner et al. demonstrated that AKAP proteins contribute to cocaine relapse vulnerability by endorsing amplified α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) receptor surface expression in the NAc.

Another point to highlight in the general study of cocaine addiction is the time it takes for the drug effects to wear off. This topic was evaluated in a study by Lull et al. who assessed proteomic changes in the mPFC of rats receiving cocaine for 10 days, followed by a comparison between naïve rats and rats with one day and 100-day cocaine abstinence [22]. They found that there are some protein changes due to drug administration that persist up to the 100 days of abstinence but other proteins do not. These short-term up-regulated proteins include synapse-associated proteins that are involved in vesicle exocytosis, dynamin-1, alpha-internexin and neurofilament medium. Even though the up-regulation does not persist, the effect of these proteins' increased expression may last and can be translated into persistent neuronal remodeling even after the protein level goes back to normal. It was also found that there are some protein changes that are not due to drug administration but take place during abstinence. Like most studies, the proteins involved were mostly related to metabolism and cell structure [22].

2.3. Neuroproteomics in non-human primate models

One way in which iTRAQ method was made even more efficient was in an experiment done on the orbitofrontal cortex (OFC) of non-human primates (14 days of chronic administration of 2 or 4 mg/kg of cocaine per day) [31]. In those experiments Olausson et al. isolated the synaptic compartment of the OFC: synaptoneurosomes, making the preparation less complex and more focused on synaptic proteins and function. Through 2D-DIGE and iTRAQ, they confirmed their previous findings on changes in alpha actin, beta tubulin, HSP70, trisphosphate isomerase, ATP synthase and glyceraldehyde-3-phosphate dehydrogenase, and other proteins. Similar to other studies, Olausson et al. found that there was a change in the expression of proteins related to cytoskeletal, metabolic and signaling functions. Importantly, this study showed that animal models other than rodents could be employed in proteomic studies.

Another study by Tannu et al. performed on rhesus monkeys examined the effect of chronic cocaine administration (0.5 mg/kg intra-venous (IV)) on protein expression and phosphorylation in the NAc via 2D-DIGE and 2DGE/phosphoprotein gel staining, respectively [42]. The latter were also analyzed by MALDI TOF/TOF MS. The proteins identified here were also related to metabolism/mitochondrial function, cell structure, synaptic plasticity and signal transduction.

Those proteins included the up-regulated glial fibrillary acidic protein (GFAP), protein kinase C isoform, syntaxin binding protein 3, mitochondrial-related proteins and adenylate kinase isoenzyme 5, and the down-regulated neural and non-neural specific enolase and beta-soluble N-ethylmaleimide-sensitive factor attachment protein. Furthermore, 15 phosphoproteins were also identified in the NAc, indicating their importance in the functional proteome. The study also revealed an increased expression of brain-type aldolase, GABA-A receptor associated protein 1, glutathione S-transferase and 14-3-3 gamma protein, and a decreased expression of PRDX2 isoform b, beta actin, guanine deaminase, Rab GDP dissociation inhibitor and other mitochondrial proteins [42].

2.4. Neuroproteomics in post-mortem human samples

In order to detect the proteomic changes that take place in the human brain more accurately, one study examined the effect of chronic cocaine abuse on the human NAc, using 2D-DIGE [43]. Tannu et al. extracted the NAc from frozen brain tissue of 10 cocaine overdose (COD) abusers and 10 controls, and only analyzed the cytosolic fractions by MALDI TOF/TOF MS. Out of the 1407 spots that were found, 18 spots were differentially expressed and were identified as 15 proteins via peptide mass fingerprinting (PMF). One of the up-regulated protein was liprin α3, a postsynaptically expressed protein, responsible for transporting AMPA and N-methyl-D-aspartate (NMDA) receptor vesicles along microtubules, which in turn have a role in mediating cocaine relapse in the NAc [43]. Some of the differentially regulated proteins found by Tannu et al. included PRDX2, which was shown to be a neuronal H_2O_2 regulator [80], neural specific enolase, ATP synthase beta chain, and the cell structure proteins TUB2, desmoplakin, GFAP and chaperonin. In addition, parvalbumin, which was shown to regulate intracellular calcium [81], was shown to be down-regulated in the study by Tannu et al. Moreover, the change in protein expression may be either due to cocaine itself or due to the up-regulated GluR receptors, which may increase calcium flux and cause oxidative stress [43].

Even though the different models of cocaine usage included different species, there were still some common differentially expressed proteins among them. These proteins include ATP synthase, PRDX2, actin, tubulin, HSP, GFAP, kinases, neural specific enolase and glutathione S-transferase. This overlap among different species suggests that the signaling pathways induced by cocaine are quite similar among different organisms and this suggests that we are able to have an idea about the effect of cocaine on the human brain by finding the effect on animal brains. Moreover, even though there were proteins that did not overlap between all organisms, the affected proteins in all models belonged to the same fields such as metabolism, cell structure and cell signaling.

3. Methamphetamine neuroproteomics

3.1. General overview

Methamphetamine belongs to the amphetamine family of drugs and is one of the most abused drugs worldwide [82] with an estimated number of 440,000 abusers in the US alone (SAMHSA, 2013). Repeated exposure to METH leads to sensitization, which is an increased behavioral response to subsequent exposures [83] due to chronically altered neuronal plasticity [44]. The negative consequences of METH abuse have been extensively studied with its most prominent effect being the destruction of dopaminergic neurons that project from the substantia nigra to the striatum [84]. High dosages of METH may lead to fatal seizures, cerebrovascular hemorrhages, hyperthermia and death [85] while withdrawal symptoms include depression, psychomotor dysfunctions and psychosis [86]. The major neuropsychiatric consequences include memory problems, motor skills and attention deficits and poor decision-making, which may be attributed to the destruction of the dopaminergic fronto-striato-cortical neurons [87]. METH is so potent that it can cross the blood–brain-barrier and the placental barrier and cause long-term effects on the brain and fetal development [88].

METH induces neuropathological changes in the brain through several mechanisms. These include decreasing the dopamine transporter (DAT) level in the dorsolateral prefrontal cortex, orbitofrontal cortex, caudate putamen [89] and basal ganglia [90]. Furthermore, DA is thought to be released from storage vesicles into the cytoplasm and extracellular matrix, leading to the production of reactive oxygen species [44] and quinine by-products, which are associated with lipid peroxidation at the monoaminergic terminals [91]. METH administration can also induce neurotoxicity at the glutamatergic terminals [92] and in different brain regions such as the striatum, parietal cortex, neocortex and somatosensory cortex [93]. Decrease in cortical gray matter [94] and serotonin transporters of orbitofrontal and cingulate cortices have also been documented [95]. METH abusers also have a decreased level of the neuronal marker N-acetylaspartate and an increased level of the glial marker myoinositol [96] as well as increased microglial activation [97]. METH can also induce apoptotic and necrotic pathways via glutamate release, leading to nitric oxide formation [98] and calcium-dependent calpain activation [99]. In addition to that, METH can induce the activation of caspase-3, 9 and 12 and the increase in expression of GRP78/BiP and of C/Ebp homologous protein that are involved in apoptosis [100]. Likewise, it also up-regulates the Bax, Bad and Bid pro-apoptotic genes and down-regulates the Bcl-2 and Bcl-xL anti-apoptotic genes [101].

Like cocaine, there are many studies that investigated proteomic changes in different brain regions and at different dosages, as a result of METH administration. Also, it has been shown that METH prompts changes in proteins associated with metabolism/mitochondrial function, cytoskeleton, synaptic plasticity, cell signaling and protein degradation.

3.2. Neuroproteomics on rat striatum

Iwazaki et al. investigated the effect of a single, low dose (1 mg/kg ip) of METH on protein expression in the striatum of rats [44]. Using 2DE-based proteomics they found 36 differentially expressed proteins in the striatum, of which 26 were increased and 10 decreased in METH-treated rats compared to saline-treated rats. When METH enters the mitochondria, it decreases ATP-synthase activity and thus raises the pH of the inner membrane matrix and reduces the membrane potential control, which disrupts normal cellular functioning [44]. The proteins related to mitochondrial function that were differentially expressed included: phosphoglycerate kinase 1, pyruvate kinase, ATP synthase *F*(1) beta chain, alpha enolase, isocitrate dehydrogenase [NAD] subunit alpha, DLP1 splice variant 3 (related to apoptosis), cystatin beta (related to apoptosis), creatine kinase and lactate dehydrogenase that are markers of cellular injury. In addition to those proteins, Iwazaki et al. found that there was an increase in dihydrolipoamide dehydrogenase, an enzyme involved in the Kreb's cycle, age-related activity and oxidative inhibition [102], and voltage-dependent anion-selective channel protein 1, which is involved in mitochondrial pore formation to release cytochrome C in apoptosis [103]. As for the metabolic-related proteins that were differentially expressed in the striatum, there were PRDX 2 and 5 and the glucose-regulated protein precursor. Moreover, the proteins related to the ubiquitin–proteasome pathway included: proteasome RN3 subunit, ubiquinol-cytochrome C reductase core protein I and ubiquitin carboxy-terminal hydrolase L1 (UCH-L1). UCH-L1 was found to be involved in germ cell and neuronal apoptosis [104] and in the degenerative process of Alzheimer's and Parkinson's diseases [105,106]. Finally, Iwazaki et al. also found that there was a change in

the expression of N-tropomodulin, GDI alpha and beta actin, indicating cytoskeletal modification without morphological changes even at a single low, non-toxic level of METH administration (1 mg/kg) [44].

Iwazaki et al. performed another study to determine the proteomic profile of rat striatum following METH-induced behavioral sensitization (2 day saline injection, followed by 1 mg/kg METH on 3rd day and then 4 mg/kg for 10 days ip) [45]. Using MALDI-TOF MS, they found 42 differentially expressed proteins, out of which 31 were identified. Some of the identified proteins include synapsin II, hippocalcin, DRP2, PEBP, adenylyl cyclase-associated protein 1 (CAP1) and synaptosomal-associated protein 25 (SNAP-25). Studies suggest that the synapsin family can modulate vesicle trafficking [107] and aide in docking the vesicles at the active zone [108]. Moreover, hippocalcin is a neuronal calcium sensor and it has been shown to be neuroprotective against calcium-induced cell death [109].

A noteworthy work by Liao et al. studied the effects of METH-induced nigrostriatal dopaminergic neurotoxicity in a mouse model (3 injections of 10 mg/kg via infusion cannula aimed at the striatum and substantia nigra) [110]. Similar to previous studies, α-synuclein, F-actin capping protein B subunit, ATPase, peroxidase and ubiquitin UCH-L1 were shown to be differentially expressed [110].

3.3. Neuroproteomics on rat amygdala

Another study by Iwazaki et al. studied the effect of acute and chronic METH administration on the amygdala of rats (acute: ip saline first 2 days, 1 mg/kg on day 3; chronic: ip saline first 2 days, 1 mg/kg on day 3, 4 mg/kg from day 4 to 14 and 1 mg/kg on day 22) [46]. Using MALDI-TOF, 43 and 48 proteins were identified in METH sensitized rats and rats with acute METH administration, respectively. Interestingly, they found that there were only three differentially expressed proteins in common between the amygdala and the striatum, and those were TP1 protein, synapsin-2b and tropomyosin. This means that these two brain regions react differently to METH administration. Intriguingly, chronic and acute METH administrations created two different patterns of protein level in the amygdala with an overlap of only nine proteins. Those proteins were LDH-B, Tpi1, Gapd protein, phosphoglycerate mutase B, fructose-bisphosphate aldolase C, glutathione S-transferase (GSTP), synapsin-2b and PRDX 5 and 6. But all in all, with all these variations, the identified proteins were mostly involved in metabolism, cellular cytoskeleton, oxidative stress and apoptosis [46].

3.4. Neuroproteomics on multiple rat brain regions and traumatic brain injury

In 2008, Li et al. studied the proteomic profile of the hippocampus, striatum and frontal cortex (8 ip injections of 15 mg/kg at 12 h intervals) [47]. They found 14, 12 and 4 differentially expressed proteins in the striatum, hippocampus and frontal cortex, respectively. Even though each brain region had a different pattern of protein change, two or three different brain regions shared some proteins. Herein, these brain regions may have followed similar pathways of METH-induced neurotoxicity, different from those followed by the amygdala. The common proteins were ATP synthase D chain, α-synuclein, mitochondrial Cu−Zn superoxide dismutase (SOD1), stathmin 1, ubiquitin-conjugating enzyme E2N, and DDAH1. It is thought that α-synuclein, E2N and SOD1 may have been up-regulated as a protective mechanism [47]. Indeed, α-synuclein has been found to interact with DA-derived oxidative species to prevent oxidative damage [111], while E2N has been found to be involved in degrading the harmful nitrated and oxidatively modified proteins that result from METH administration [47]. As for the SOD1, it can scavenge for exogenous and endogenous superoxide ions [112].

Even though the exact mechanism by which METH induces an increase in nitric oxide production is unknown [113], one hypothesis is that METH induces the activation of the DDAH1/ ADMA (asymmetric dimethylated L-arginine)/NOS (nitric oxide synthase) pathway [48]. In a recent study, Zhang et al. found that administering a DDAH1 inhibitor, Nx-(2-methoxyethyl)-arginine (L-257), to METH-treated rats would decrease the nitric oxide production, meaning that METH acts through the DDAH1/ADMA/NOS pathway [48]. In this same study, it was found that there was an increase in 30 nitrated proteins, including tropomodulin 1 and 2, in both rats and PC12 cells (rats: 8 ip injections of 15 mg/kg at 12 h intervals; PC12: 2 mM). PC12 are pheochromocytoma cells derived from the rat adrenal medulla, and can develop characteristics of catecholaminergic sympathetic neurons when treated with nerve growth factor [114]. Stable isotope labeling with amino acids in cell culture (SILAC) was employed for detecting nitrated proteins in METH-treated PC12 because, according to Zhang et al., 2-DE is not as efficient for that job. Zhang et al. were interested in the increased expression of the nitrated protein GSTP1 and the decreased expression of the normal GSTP1, which is known for its protective role against oxidative stress. The nitration of GSTP1 has been shown to subsequently activate the p25/p35/cdk5 pathway which will in turn impose oxidative stress [115]. Taking that into consideration, Zhang et al. elucidated a potential mechanism through which METH can induce an increase in NO production via the DDAH1/ ADMA/NOS pathway and the p25/p35/cdk5 pathway.

The deleterious effects of METH on the CNS have also been compared to TBI. In a study by Warren et al., the effect of acute ip METH administration (10, 20 or 40 mg/kg) on rat hippocampus and cortex was analyzed and compared to the changes following TBI [49]. Using SDS-PAGE, Warren et al. found that there was an increase in the breakdown products of MAP-τ and αII-spectrin (SBDP120 and SBDP150). The presence of these products suggests neuronal cell death [116,117]. Warren et al. found that the severity of neurotoxicity was almost equal to TBI when a dosage of 40 mg/kg of METH was administered. They suggested that a single METH administration mediates neurotoxicity via calpain-induced proteolysis in the cortex, and calpain and caspase-induced proteolysis in the hippocampus, leading to cytoskeletal changes in neurons [49].

After acute METH administration (ip 40 mg/kg), Kobeissy et al. found 82 differentially expressed proteins in the rat cortex, of which 42 were up-regulated and 40 were down-regulated [50]. The identified proteins were involved in oxidative stress, synaptic transmission and cell death. The up-regulated proteins included UCH-L1, UCH-L3, synapsin I protein and microtubule-associated protein light chain 3 (LC3). LC3 is attached to autophagosomes [118], so in other words, METH is inducing the formation of autophagic vesicles. These vesicles are able to engulf oxidized and degraded proteins that may kill the neurons [119]. On the other hand, down-regulated proteins that Kobeissy et al. found included SOD1, MKK1, PEBP1, collapsin response mediator protein 2 (CRMP2) and the F-actin scaffolding protein that has been proven to be a neuronal cytoskeleton stabilizer [110]. Both PEBP1 and CRMP2 were found to be substrates of the calpain pathway [120,121], and since METH acts through this pathway, it accounts for the decreased level of these substrates. Kobeissy et al. concluded that there was a similarity in the proteomic changes taking place after both TBI and acute METH treatment, meaning that there could be common neurobiological events leading to the long-term neurological and neuropsychiatric effects [50].

Additionally, Yang et al. found that a positive CPP test was established after three discontinuous (ip) injections of 1 mg/kg of METH in rats [51]. Protein profiles in the NAc, PFC, cingulate cortex, striatum and hippocampus were then compared. They identified 27 differentially expressed proteins that were related to the same functions as the proteins found by other studies. Proteins related to the cytoskeleton included LMOa7, G protein b-subunit, neurofilament and Neogenin; while the proteins related to cell signaling included N-myc downstream-regulated gene 2, G protein b-subunit soluble guanylyl cyclase, GDP dissociation inhibitor 1 and 14-3-3 protein. The 14-3-3

protein is functionally important in altering behavior following subchronic METH treatment, activating protein kinase C (PKC), Raf-1 in the signal transduction mitogen activated protein kinase (MAPK) cascade, and tryptophan and tyrosine hydroxylases [122].

In addition, it has also been shown that PKC is an important mediator of behavioral sensitization to METH [123]. In the study of Yang et al., the expression of the 14-3-3 protein was decreased in the hippocampus only. So, there exists a discrepancy where Kobeissy et al. found a decrease in PEBP1 in the rat cortex while Yang et al. found a decrease in the 14-3-3 protein, which is a Raf activator, in the hippocampus. This means that the cortex may be activating the PKC pathway, thus augmenting behavioral sensitization, while the hippocampus is doing the exact opposite. Identified proteins related to transport, endocytosis and exocytosis included profilin-2 and syntaxin. Yang et al. suggested that the alteration of Profilin-2 expression might lead to increased post-synaptic endocytosis in the striatum and NAc, thus, inducing a decrease in intracellular calcium and dopamine D2 receptor internalization [51]. Interestingly, intracellular calcium is involved in developing sensitization [124], so there is another discrepancy here where a decrease in intracellular calcium would mean that there is an attenuation of behavioral sensitization to METH. This discrepancy is further maintained in a study by Borlongan et al. that showed that decreasing intracellular calcium via cyclosporine-A would increase METH-induced locomotor behavior [125].

In addition to using different models, drugs and dosages and examining different brain regions, one should also take into consideration identifying proteins as cytosolic or membrane bound because these factors may influence the proteomic profile. Unfortunately, there is only one study that categorized proteins into cytosolic and membrane bound. In this 2009 study, Faure et al. examined the proteomic profiles of rat frontal cortex following eight METH injections (ip, 1 mg/kg) [52]. There were 47 cytosolic and 42 membrane proteins that were differentially expressed, which were associated with metabolism, redox regulation, protein degradation, cellular growth, synaptic function and cytoskeletal modifications.

3.5. Neuroproteomics on rat striatum and olfactory bulb

In a recent study from our lab, escalating chronic model of methamphetamine was assessed for differential proteomic profile in two rat brain regions: the olfactory bulb and the hippocampus (data not published). METH was injected (ip) for 2 weeks, starting from 0.5 mg/kg and ending with 8 mg/kg on the 14th day. To identify differentially expressed proteins, we applied LC–ESI-MS/MS, followed by multiple reaction monitoring (MRM) LC–MS/MS for confirmation. There were 11 differentially expressed proteins in the hippocampus, including the down-regulated spectrin alpha chain non-erythrocytic 1 (SPTN1) and the up-regulated synaptic vesicle glycoprotein 2A (SV2A), as well as the myelin proteolipid protein (MYPR), brain acid soluble protein 1 (BASP1), V-type proton ATPase subunit B brain isoform (VATB2) and glutathione S-transferase alpha 3 (GSTA3). Moreover, we found seven differentially expressed proteins in the olfactory bulb, including the down-regulated alpha-2-HS-glycoprotein (FETUA) and the up-regulated 6-phosphofructokinase type C (K6PP), as well as the CNRIP1, Secernin-1 (SCRN1), and the guanine nucleotide-binding protein G(i) subunit alpha 1 and 2 (GNAI1 and GNAI2). SV2A is involved in regulating synaptic vesicle transport [126] but not in the release, storage and uptake of neurotransmitters [127]. According to Hanaya et al., decrease in SV2A levels may interfere in GABAergic functions and induce epileptic seizures and sclerotic changes throughout the hippocampus [128]. On the other hand, non-erythroid spectrin is involved in regulating neuritogenesis [129], assembling the Nodes of Ranvier and stabilizing sodium channels [130]. GNAI is involved in intracellular signaling, where the alpha subunit is specifically responsible for transiently binding GTP and GDP [131]. Furthermore, FETUA was found to be structurally similar to TGF-B receptor [132] and was able to interfere with insulin receptor signaling [133]. Moreover, secernin-1, was found to play an integral role in regulating exocytosis from peritoneal mast cells and affecting mast cell sensitivity to stimulation with calcium [134]. Secernin-1 has also been implicated as a biomarker for gastric and colon cancer [135], and more recently, synovial sarcoma [136].

Neurosystems biology analysis of differential proteins was performed utilizing PANTHER (protein analysis through evolutionary relationships) software and Pathway Studio software to construct an interaction map, exhibiting direct and indirect pathways of proteins, which highlighted the involvement of metabolic, neuroprotective, cytoskeletal, cell signaling and transport pathways as shown in Fig. 2. In this work, 35 differential proteins were identified in the hippocampus brain region (20 down-regulated vs. 15 up-regulated proteins) while (9 down-regulated vs. 13 up-regulated proteins) in the olfactory bulb (data presentation pending publishing). A subset of these proteins were validated in both brain regions and were used to assess the protein interaction map and the biological process involved, as shown in Fig. 2.

4. MDMA neuroproteomics

4.1. General overview

Today, the abusive intake of 3,4-methylenedioxymethamphetamine, also known as MDMA or more commonly known as Ecstasy, is an escalating issue both in the US and many parts of the world, making it a public health problem [137–139]. MDMA usage has been decreasing globally, except for Europe that has witnessed an increase to reach a number of about 2 million users in the past year. The acute exposure to MDMA is known to cause damage to organs such as brain, heart, liver, and kidneys. Such damages can be deadly, causing myocardial infarction usually accompanied with hyperthermia, hypertension, and tachycardia, which precede disseminated intravascular coagulation, rhabdomyolysis, multiple organ failure and, eventually, death [140]. When administered to humans and non-humans, MDMA damaged serotonin cell bodies and nerve terminals [141] and induced neuronal loss in the nigrostriatal dopaminergic terminals and the substantia nigra compacta [142]. MDMA also increased the reactive oxygen species in the monoaminergic systems [143] and decreased vesicular DA transport [144]. Not to mention that MDMA metabolism produced 3,4-methylenedioxyamphetamine (MDA), 3,4-dihydroxyamphetamine (HHA) and 3,4-dihydroxymethamphetamine (HHMA) that can form thioether metabolites or quinone compounds, which produce free radicals [141].

Although many studies have proven that the use of MDMA does, in fact, cause liver damage, the underlying mechanisms were unknown until recently. Many studies in the field of proteomics, such as those done by Moon et al. and Upreti et al., have touched upon the mechanism behind the induced damage [57,58]. Using comparative 2-DE in the analysis of the proteins in the liver that undergo modification after acute MDMA administration, they were able to have an idea about the mechanisms underlying liver damage. The studies mentioned below aim to explain the toxicity mechanisms of MDMA, including those found in liver tissue. It is believed that the mechanisms taking place in hepatocytes also apply to other cells and tissues such as the brain and kidney [57].

4.2. Proteomic studies on the liver

After orally administering 10 mg/kg of MDMA on day 1 and 2, Moon et al. and Upreti et al. observed a great difference between the protein expression in controls and MDMA-exposed rat livers. There were around 33 proteins identified by Moon et al. and around 22 by

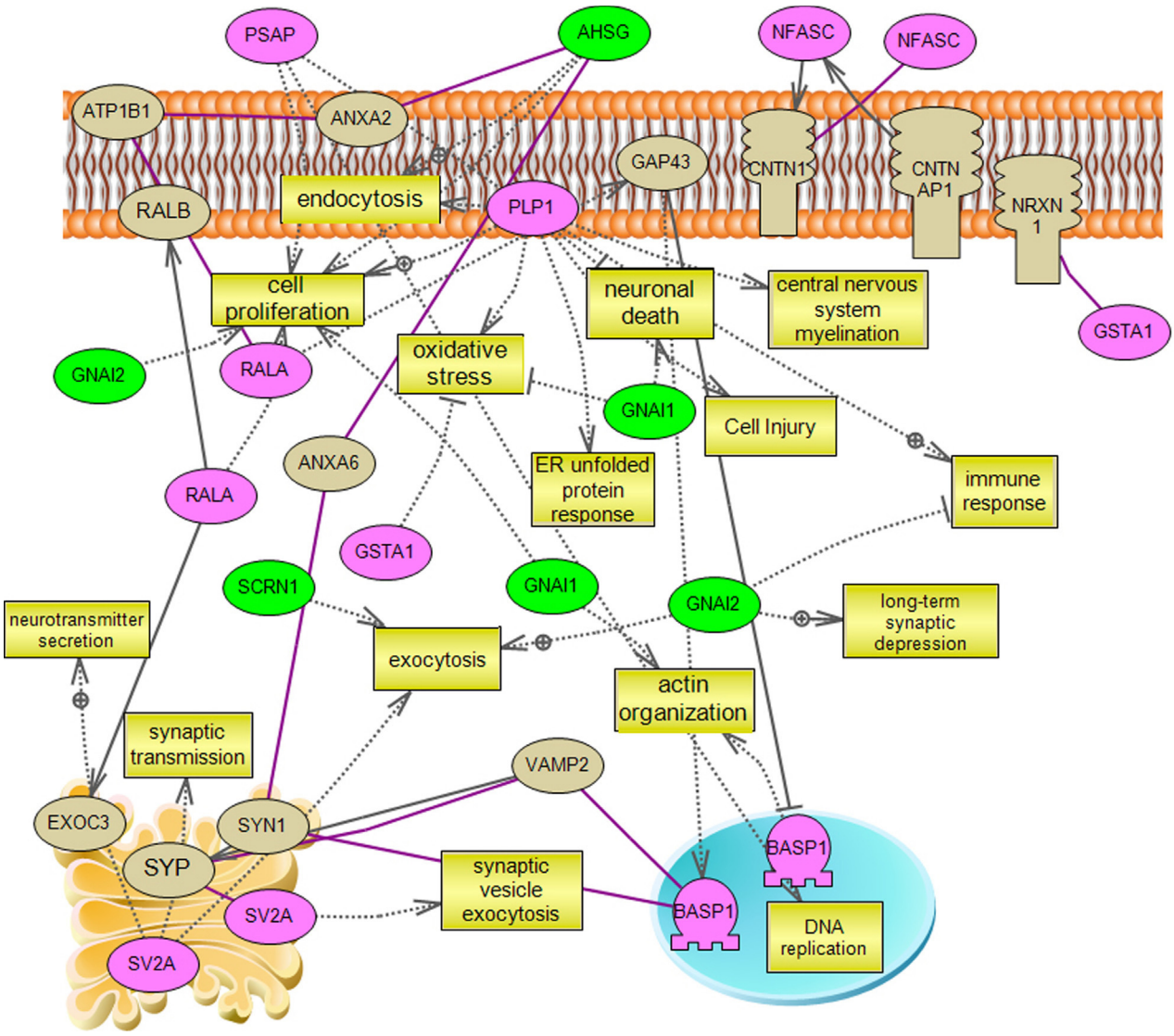

Fig. 2. Global interaction proteome: associating biological processes and altered METH-induced proteome. The altered nodes (genes) and edges (gene relationships) are described in the figure. The node color indicates gene regulation where purple nodes reflect up-regulation while green nodes reflect down-regulation. The gray-colored genes are not identified as differentially expressed genes in our experiment but are integrated into the computationally generated networks on the basis of third party connectors that highlight the relation to the identified network. Furthermore, yellow boxes indicate identified biological processes. The pathway includes interacting proteins of metabolism, neuroprotection, cytoskeleton, cell signaling and transport.

Upreti et al. The 22 differentially expressed proteins identified by Upreti et al. were oxidatively modified proteins belonging to the domains of metabolism, calcium-binding proteins and antioxidant defensive enzymes [58]. The 33 differentially expressed proteins identified by Moon et al. belonged to the domains of energy supply, oxidative phosphorylation, mitochondrial electron transport, anti-oxidative defense system, fatty acid and intermediary metabolism, molecular chaperon proteins and voltage-dependent calcium proteins [57]. Moon et al. identified other spots as fragments of full-length proteins; such spots represented proteins of smaller molecular weight (e.g. 26–30 kDa). The fragmentation of these proteins may have been due to either the increased susceptibility of mitochondrial proteins to proteasomal degradations under oxidative stress [145,146] or simply the spontaneous fragmentation of oxidized proteins [147].

It is the quinone metabolites of MDMA, produced via cytochrome P450-catalyzed metabolism, that are important in MDMA-induced organ damage, while the direct administration of MDMA to brain tissue does not cause any tissue damage [148,149]. The main reason for this is that these metabolites produce oxidative and nitrosative stress by the inhibition of the mitochondrial respiratory chain [150–152]. However, the way these metabolites work on the suppression of the mitochondrial respiratory chain is still unknown. In addition, MDMA metabolites work on decreasing the level of reduced glutathione, which is protective against a broad range of metabolite-related free radicals of several toxic compounds. The presence of both reactive oxygen and reactive nitrogen species can create a more potent oxidant peroxynitrite ($ONOO^-$), which may be the cause of irreversible tissue and cell damage [153].

More mitochondrial proteins such as acetyl-CoA dehydrogenase, UDP-glucose pyrophosphorylase 2, HSP86, HSP75, and cytochrome P450 11B1 (CYP11B1) were also found [57]. Moon et al. showed oxidative patterns of both CYP11B1 and CYP19A1, however there was no oxidation of CYP2D6, CYP1A2, and CYP3A4, which are involved in MDMA metabolism [154]. This is most probably because of their low expression levels [57]. Moon et al. and Upreti et al. have speculated that the mechanism behind protein inactivation by MDMA is due to oxidative stress caused by the metabolites of MDMA, and the direct binding of metabolites to these proteins [57,58].

4.3. Neuroproteomic studies on the brain

In a study performed by Van Nieuwenhuijzen on rat hippocampus, animals were chronically administered MDMA for 10 days (ip 5 mg/kg) and were finally sacrificed 8 weeks post-administration [54]. MDMA administration provoked residual changes in eight proteins involved in intracellular signaling (C-crk, 14-3-3 protein epsilon and adenyl cyclase), neuroplasticity (neuronal pentraxin-1), transport (alpha-soluble NSF-attachment protein and V-type proton ATPase subunit F) and cytoskeletal function (tubulin folding cofactor B and tropomyosin alpha 3 chain).

In another study by Puerta et al., the administration of an increasing dose of MDMA (10, 20, 30 mg/kg (ip) injected every 2 h) resulted in a noticeable decline in mitochondrial complex I activity [55]. MDMA-treated mice showed production of O_2- or O_2-derived oxidants in their corpus striatum, 3 h following MDMA treatment. After 3 and 6 h of MDMA exposure, a noticeable decline in aconitase activity (metabolic function) was also noted. The study found that administration of NG-nitro-l-arginine (l-NNA), a nitric oxide synthase inhibitor, has significantly served as a neuro-protective agent against DA loss [55]. Moreover, the addition of lipoic acid inhibited the formation of reactive oxygen species but did not reverse the MDMA-induced inhibition of complex I. This suggests that a complex I inhibition happens early in MDMA neurotoxicity. Additionally, Puerta et al. treated the mice with MDMA and the Parkinsonian toxin 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP). They co-administered the non-toxic MDMA treatments and it resulted in a substantial long-term decline of striatal DA concentration. However, the addition of MPTP did not result in increasing the effect of MDMA, and thus they concluded that DA depletion was the result of MDMA alone [55].

In a second study, researchers aimed at finding the effects of MDMA on the hippocampus of DSP-4/MDMA-treated rats and mice (ip injection of 100 mg/kg DSP-4 and 4 sc injection of 10 mg/kg MDMA) [56]. DSP-4 is a neurotoxin that affects noradrenergic neurons by depleting their noradrenaline [155]. After 2 weeks of continuous MDMA exposure, Biezonski and Meyer found no significant changes in the expression levels of vesicular monoamine transporter 2 protein (VMAT-2) in MDMA-treated animals compared to DSP-4/saline controls. However, serotonin transporter (SERT) levels were substantially declined in all MDMA-treated groups. This means that although amounts of SERT did decline, there was no terminal induced loss [56]. Furthermore, Biezonski and Meyer found a declined gene expression of SERT and VMAT-2, showing that DSP-4 may regulate serotonergic neurons without affecting distal axotomy.

Finally, unlike the case of cocaine and METH, there are only a few studies that were done to assess proteomic profile changes post MDMA exposure. Of interest are the MDMA studies that focused on MDMA distribution throughout the body [156] and its effects on different organs other than the brain [157–159]. Even though the number of studies on the effect of MDMA on the brain is negligible, the effect of MDMA on other organ systems may still give us a clue about what is happening in the brain.

5. Conclusion

To sum up, the aim behind proteomic studies is to assess the effect of drugs on humans and try to find therapeutic methods to treat addiction and abuse. Unfortunately, there are only few proteomic studies on drug abuse. Moreover, most studies have used animal models to profile protein expression following drug administration; but protein expression in animals might be different from protein expression in humans. Moreover, few studies have been conducted on postmortem human samples, whether drug abusers or controls, and they might not be very reliable in representing drug effects in living humans; that is because the people from whom the samples were taken might have had a certain neurological or neurodegenerative disease that could have affected the protein expression in the brain; however, this situation is not exclusive to neuroproteomics studies on drug abuse models. Moreover, the cause of death in those people was not mentioned in several studies; those people may have had a TBI for example, or any other malady that might have directly or indirectly caused their death and in turn affected their brain proteomic profile much more than the drugs that they were taking. In some studies, drug overdose was mentioned as the cause of death; however, overdose may lead to different direct causes of death such as a stroke or heart attack or any other organ failure. Accordingly, the altered brain proteomic profile could be more due to the direct cause of death and less due to the effect of the drug on the brain.

Additionally, even though cocaine, METH and MDMA are chemically and structurally different, they appeared to induce common pathways and cause similar cellular changes. The pathways were mainly involved in metabolism/mitochondrial function, cell signaling, cytoskeleton, protein modification and degradation, cell death and synaptic plasticity. Unfortunately, all of these studies were not able to detect proteins that are vital for drug addiction such as GTPases, phosphatases, kinases and neurotransmitter receptors because they are too scarce and hard to separate when compared to the other abundant proteins that are being detected. Relevant receptors such as GABA, DA and glutamate receptors were not detected in these proteomic studies even though they play an essential role in mediating the effects of drugs at the genetic and proteomic level.

Even though many studies used the same experimental design and analysis method, each got a different protein expression profile. This could be attributed to different experimental methodologies and models, experimental errors, instrumentation and insensitive MS readings. This necessitates further studies to ascertain that the differential identified proteins are truly involved in mediating the effects of drug abuse. Thus, better model selection and improved sample preparation may also be needed for detecting a wider range of proteins.

The realm of proteomics is still in its beginning and it has started to bring about many possibilities and even more questions to answer in many domains. This field could be further incorporated into the clinical setting where the level of drug addiction and the severity of drug-induced damage could be easily assessed. Nowadays, medicine is heading towards a point of care system where we are trying to develop handheld devices (such as those that measure blood glucose in diabetic patients) that are both fast and accurate in measuring blood-borne substances that help in assessing the clinical situation of a patient. We may not be far from finally attaining such a neuroproteomic handheld device to measure the level of expression of certain neuronal proteins (in the blood, CSF or maybe even brain tissue) and in turn assess the effect of any drug on the brain of the patient at hand. Yet, what remains to do is to further study the intricate brain functions and delve into the most specific cell signaling pathways that are directly and indirectly being affected by the drug in question. Only then would we understand the true effect of each drug on the brain and target the right protein or proteomic expression profile in our quest of assessing the drug's effect on the brain and accordingly establish new therapeutic modalities.

Conflict of interest

None.

References

[1] Wu L.T., Blazer D.G. Substance use disorders and psychiatric comorbidity in mid and later life: a review. International Journal of Epidemiology 2014;43:304–17. http://dx.doi.org/10.1093/ije/dyt173, 24163278.

[2] Mewes H.W., Wachinger B., Stumpflen V. Perspectives of a systems biology of the synapse: how to transform an indefinite data space into a model? Pharmacopsychiatry 2010;43:S2–S8. http://dx.doi.org/10.1055/s-0030-1249666, 20480444.

[3] Gold M.S., Kobeissy F.H., Wang K.K., Merlo L.J., Bruijnzeel A.W., Krasnova I.N. et al. Methamphetamine- and trauma-induced brain injuries: comparative cellular and molecular neurobiological substrates. Biological Psychiatry 2009;66:118–27. http://dx.doi.org/10.1016/j.biopsych.2009.02.021, 19345341.

[4] Westerhoff H., Kell D. The methodology of systems biology.Systems biology: philosophical foundations. Elsevier; 2007.

[5] Lange R., Iverson G., Franzen M. Comparability of neuropsychological test profiles in patients with chronic substance abuse and mild traumatic brain injury. Clinical Neuropsychologist 2008;22:209–27. http://dx.doi.org/10.1080/13854040701290062, 17853134.

[6] Albright J.C., Goering A.W., Doroghazi J.R., Metcalf W.W., Kelleher N.L. Strain-specific proteogenomics accelerates the discovery of natural products via their biosynthetic pathways. Journal of Industrial Microbiology & Biotechnology 2014;41:451–9. http://dx.doi.org/10.1007/s10295-013-1373-4, 24242000.

[7] Ramakrishnan S.R., Vogel C., Prince J.T., Li Z., Penalva L.O., Myers M. et al. Integrating shotgun proteomics and mRNA expression data to improve protein identification. Bioinformatics (Oxford, England) 2009;25:1397–403. http://dx.doi.org/10.1093/bioinformatics/btp168, 19318424.

[8] Taurines R., Dudley E., Grassl J., Warnke A., Gerlach M., Coogan A.N. et al. Proteomic research in psychiatry. Journal of Psychopharmacology (Oxford, England) 2011;25:151–96. http://dx.doi.org/10.1177/0269881109106931, 20142298.

[9] Baker E., Liu T., Petyuk V., Burnum-Johnson K., Ibrahim Y., Anderson G. et al. Mass spectrometry for translational proteomics: progress and clinical implications. Genome Medicine 2012;4:63. http://dx.doi.org/10.1186/gm364, 22943415.

[10] Moron J.A., Devi L.A. Use of proteomics for the identification of novel drug targets in brain diseases. Journal of Neurochemistry 2007;102:306–15. http://dx.doi.org/10.1111/j.1471-4159.2007.04536.x, 17419802.

[11] Anderson L., Seilhamer J. A comparison of selected mRNA and protein abundances in human liver. Electrophoresis 1997;18:533–7. http://dx.doi.org/10.1002/elps.1150180333, 9150937.

[12] Hemby S.E., Tannu N. Modeling substance abuse for applications in proteomics. Methods in Molecular Biology (Clifton, N.J.) 2009;566:69–83. http://dx.doi.org/10.1007/978-1-59745-562-6_5, 20058165.

[13] Karasawa R. Progress and clinical applications in proteomics. Journal of Data Mining in Genomics & Proteomics 2012. http://dx.doi.org/10.4172/2153-0602.

[14] Aebersold R., Mann M. Mass spectrometry-based proteomics. Nature 2003;422:198–207. http://dx.doi.org/10.1038/nature01511, 12634793.

[15] Boutet E., Lieberherr D., Tognolli M., Schneider M., Bairoch A. UniProtKB/Swiss-Prot. Methods in Molecular Biology (Clifton, N.J.) 2007;406:89–112, 18287689.

[16] Glessner J., Hakonarson H. Common variants in polygenic schizophrenia. Genome Biology 2009;10:236. http://dx.doi.org/10.1186/gb-2009-10-9-236, 19785721.

[17] Gronborg M., Kristiansen T.Z., Iwahori A., Chang R., Reddy R., Sato N. et al. Biomarker discovery from pancreatic cancer secretome using a differential proteomic approach. Molecular & Cellular Proteomics: MCP 2006;5:157–71, 16215274.

[18] Tacheny A., Dieu M., Arnould T., Renard P. Mass spectrometry-based identification of proteins interacting with nucleic acids. Journal of Proteomics 2013;94C:89–109, 24060998.

[19] Jensen L., Kuhn M., Chaffron S., Creevey C., Muller J., Doerks T. et al. String 8 – a global view on proteins and their functional interactions in 630 organisms. Nucleic Acids Research 2009;37:D412–D416. http://dx.doi.org/10.1093/nar/gkn760, 18940858.

[20] Gargus J.J. Ion channel functional candidate genes in multigenic neuropsychiatric disease. Biological Psychiatry 2006;60:177–85. http://dx.doi.org/10.1016/j.biopsych.2005.12.008, 16497276.

[21] Viswanathan S., Unlü M., Minden J. Two-dimensional difference gel electrophoresis. Nature Protocols 2006;1:1351–8. http://dx.doi.org/10.1038/nprot.2006.234, 17406422.

[22] Lull M.E., Erwin M.S., Morgan D., Roberts D.C., Vrana K.E., Freeman W.M. Persistent proteomic alterations in the medial prefrontal cortex with abstinence from cocaine self-administration. Proteomics. Clinical Applications 2009;3:462–72. http://dx.doi.org/10.1002/prca.200800055, 20161123.

[23] Choudhary J., Grant S.G. Proteomics in postgenomic neuroscience: the end of the beginning. Nature Neuroscience 2004;7:440–5. http://dx.doi.org/10.1038/nn1240, 15114355.

[24] Ottens A.K., Kobeissy F.H., Golden E.C., Zhang Z., Haskins W.E., Chen S.S. et al. Neuroproteomics in neurotrauma. Mass Spectrometry Reviews 2006;25:380–408. http://dx.doi.org/10.1002/mas.20073, 16498609.

[25] Zhang C., Omran A.G., He F., Deng X., Wu L., Peng J. et al. Screening and identification of dynamin-1 interacting proteins in rat brain synaptosomes. Brain Research 2014;1543:17–27. http://dx.doi.org/10.1016/j.brainres.2013.10.053, 24211660.

[26] Luque-Garcia J.L., Neubert T.A. Sample preparation for serum/plasma profiling and biomarker identification by mass spectrometry. Journal of Chromatography A 2007;1153:259–76. http://dx.doi.org/10.1016/j.chroma.2006.11.054, 17166507.

[27] Issaq H.J., Veenstra T.D. The role of electrophoresis in disease biomarker discovery. Electrophoresis 2007;28:1980–8. http://dx.doi.org/10.1002/elps.200600834, 17503404.

[28] Domon B., Aebersold R. Mass spectrometry and protein analysis. Science (New York, N.Y.) 2006;312:212–17. http://dx.doi.org/10.1126/science.1124619, 16614208.

[29] Merchant M., Weinberger S.R. Recent advancements in surface-enhanced laser desorption/ionization-time of flight-mass spectrometry. Electrophoresis 2000;21:1164–77, 10786889.

[30] Issaq H.J., Veenstra T.D., Conrads T.P., Felschow D. The SELDI-TOF MS approach to proteomics: protein profiling and biomarker identification. Biochemical and Biophysical Research Communications 2002;292:587–92. http://dx.doi.org/10.1006/bbrc.2002.6678, 11922607.

[31] Olausson P., Jentsch J.D., Krueger D.D., Tronson N.C., Nairn A.C., Taylor J.R. Orbitofrontal cortex and cognitive-motivational impairments in psychostimulant addiction: evidence from experiments in the non-human primate. Annals of the New York Academy of Sciences 2007;1121:610–38. http://dx.doi.org/10.1196/annals.1401.016, 17698993.

[32] Aggarwal K., Choe L.H., Lee K.H. Shotgun proteomics using the iTRAQ isobaric tags. Briefings in Functional Genomics & Proteomics 2006;5:112–20. http://dx.doi.org/10.1093/bfgp/ell018, 16772272.

[33] Foster L.J., de Hoog C.L., Zhang Y., Zhang Y., Xie X., Mootha V.K. et al. A mammalian organelle map by protein correlation profiling. Cell 2006;125:187–99. http://dx.doi.org/10.1016/j.cell.2006.03.022, 16615899.

[34] Kislinger T., Cox B., Kannan A., Chung C., Hu P., Ignatchenko A., et al. Global survey of organ and organelle protein expression in mouse: combined proteomic and transcriptomic profiling. Cell 2006;125:173–86. http://dx.doi.org/10.1016/j.cell.2006.01.044, 16615898.

[35] Washburn M., Wolters D., Yates J.R. Large-scale analysis of the yeast proteome by multidimensional protein identification technology. Nature Biotechnology 2001;19:242–7. http://dx.doi.org/10.1038/85686, 11231557.

[36] Friedman D.B., Hill S., Keller J.W., Merchant N.B., Levy S.E., Coffey R.J. et al. Proteome analysis of human colon cancer by two-dimensional difference gel electrophoresis and mass spectrometry. Proteomics 2004;4:793–811. http://dx.doi.org/10.1002/pmic.200300635, 14997500.

[37] Byrne J.C., Downes M.R., O'Donoghue N., O'Keane C., O'Neill A., Fan Y. et al. 2D-DIGE as a strategy to identify serum markers for the progression of prostate cancer. Journal of Proteome Research 2009;8:942–57. http://dx.doi.org/10.1021/pr800570s, 19093873.

[38] Orlowska K.P., Klosowska K., Szczesny R.J., Cysewski D., Krawczyk P.S., Dziembowski A. A new strategy for gene targeting and functional proteomics using the DT40 cell line. Nucleic Acids Research 2013;41:e167. http://dx.doi.org/10.1093/nar/gkt650, 23892402.

[39] Del Castillo C., Morales L., Alguacil L.F., Salas E., Garrido E., Alonso E. et al. Proteomic analysis of the nucleus accumbens of rats with different vulnerability to cocaine addiction. Neuropharmacology 2009;57:41–8. http://dx.doi.org/10.1016/j.neuropharm.2009.04.005, 19393250.

[40] Guan X., Guan Y. Proteomic profile of differentially expressed proteins in the medial prefrontal cortex after repeated cocaine exposure. Neuroscience 2013;236:262–70. http://dx.doi.org/10.1016/j.neuroscience.2013.01.039, 23376741.

[41] Reissner K.J., Uys J.D., Schwacke J.H., Comte-Walters S., Rutherford-Bethard J.L., Dunn T.E. et al. AKAP signaling in reinstated cocaine seeking revealed by iTRAQ proteomic analysis. Journal of Neuroscience: The Official Journal of the Society for Neuroscience 2011;31:5648–58. http://dx.doi.org/10.1523/JNEUROSCI.3452-10.2011, 21490206.

[42] Tannu N.S., Howell L.L., Hemby S.E. Integrative proteomic analysis of the nucleus accumbens in rhesus monkeys following cocaine self-administration. Molecular Psychiatry 2010;15:185–203. http://dx.doi.org/10.1038/mp.2008.53, 18504425.

[43] Tannu N.S., Mash D., Hemby S. Cytosolic proteomic alterations in the nucleus accumbens of cocaine overdose victims. Molecular Psychiatry 2007;12:55–73. http://dx.doi.org/10.1038/sj.mp.4001914, 17075605.

[44] Iwazaki T., McGregor I.S., Matsumoto I. Protein expression profile in the striatum of acute methamphetamine-treated rats. Brain Research 2006;1097:19–25. http://dx.doi.org/10.1016/j.brainres.2006.04.052, 16729985.

[45] Iwazaki T., McGregor I.S., Matsumoto I. Protein expression profile in the striatum of rats with methamphetamine-induced behavioral sensitization. Proteomics 2007;7:1131–9. http://dx.doi.org/10.1002/pmic.200600595, 17351886.

[46] Iwazaki T., McGregor I.S., Matsumoto I. Protein expression profile in the amygdala of rats with methamphetamine-induced behavioral sensitization. Neuroscience Letters 2008;435:113–19. http://dx.doi.org/10.1016/j.neulet.2008.02.025, 18346852.

[47] Li X., Wang H., Qiu P., Luo H. Proteomic profiling of proteins associated with methamphetamine-induced neurotoxicity in different regions of rat brain. Neurochemistry International 2008;52:256–64. http://dx.doi.org/10.1016/j.neuint.2007.06.014, 17904249.

[48] Zhang F., Chen L., Liu C., Qiu P., Wang A., Li L. et al. Up-regulation of protein tyrosine nitration in methamphetamine-induced neurotoxicity through DDAH/ADMA/NOS pathway. Neurochemistry International 2013;62:1055–64. http://dx.doi.org/10.1016/j.neuint.2013.03.016, 23583342.

[49] Warren M.W., Kobeissy F.H., Liu M.C., Hayes R.L., Gold M.S., Wang K.K. Concurrent calpain and caspase-3 mediated proteolysis of alpha II-spectrin and tau in rat brain after methamphetamine exposure: a similar profile to traumatic brain injury. Life Sciences 2005;78:301–9. http://dx.doi.org/10.1016/j.lfs.2005.04.058, 16125733.

[50] Kobeissy F.H., Warren M.W., Ottens A.K., Sadasivan S., Zhang Z., Gold M.S. et al. Psychoproteomic analysis of rat cortex following acute methamphetamine exposure. Journal of Proteome Research 2008;7:1971–83, 18452277.

[51] Yang M.H., Kim S., Jung M.-S., Shim J.H., Ryu N.K., Yook Y.J. et al. Proteomic analysis of methamphetamine-induced reinforcement processes within the

mesolimbic dopamine system. Addiction Biology 2008;13:287–94, 18279499.
[52] Faure J.J., Hattingh S.M., Stein D.J., Daniels W.M. Proteomic analysis reveals differentially expressed proteins in the rat frontal cortex after methamphetamine treatment. Metabolic Brain Disease 2009;24:685–700. http://dx.doi.org/10.1007/s11011-009-9167-0, 19826936.
[53] Liao P.C.K., Hsu H.C., Cherng C.G., Yu L. Local proteins associated with methamphetamine-induced nigrostriatal dopaminergic neurotoxicity. Journal of Neurochemistry 2005;95:160–8. http://dx.doi.org/10.1111/j.1471-4159.2005.03346.x, 16181420.
[54] van Nieuwenhuijzen P.S., Kashem M.A., Matsumoto I., Hunt G.E., McGregor I.S. A long hangover from party drugs: residual proteomic changes in the hippocampus of rats 8 weeks after gamma-hydroxybutyrate (GHB), 3,4-methylenedioxymethamphetamine (MDMA) or their combination. Neurochemistry International 2010;56:871–7. http://dx.doi.org/10.1016/j.neuint.2010.03.002, 20227452.
[55] Puerta E., Hervias I., Goñi-Allo B., Zhang S.F., Jordán J.n., Starkov A.A. et al. Methylenedioxymethamphetamine inhibits mitochondrial complex I activity in mice: a possible mechanism underlying neurotoxicity. British Journal of Pharmacology 2010;160:233–45. http://dx.doi.org/10.1111/j.1476-5381.2010.00663.x, 20423338.
[56] Biezonski D.K., Meyer J.S. Effects of 3,4-methylenedioxymethamphetamine (MDMA) on serotonin transporter and vesicular monoamine transporter 2 protein and gene expression in rats: implications for MDMA neurotoxicity. Journal of Neurochemistry 2010;112:951–62. http://dx.doi.org/10.1111/j.1471-4159.2009.06515.x, 20002520.
[57] Moon K.H., Upreti V.V., Yu L.R., Lee I.J., Ye X., Eddington N.D. et al. Mechanism of 3,4-methylenedioxymethamphetamine (MDMA, ecstasy)-mediated mitochondrial dysfunction in rat liver. Proteomics 2008;8:3906–18. http://dx.doi.org/10.1002/pmic.200800215, 18780394.
[58] Upreti V.V., Moon K.H., Yu L.R., Lee I.J., Eddington N.D., Ye X. et al. Increased oxidative-modifications of cytosolic proteins in 3,4-methylenedioxymethamphetamine (MDMA, ecstasy)-exposed rat liver. Proteomics 2011;11:202–11. http://dx.doi.org/10.1002/pmic.201000203, 21204248.
[59] Guindalini C., O'Garaa C., Laranjeirad R., Colliera D., Castelod A., Vallada H. et al. A GSTP1 functional variant associated with cocaine dependence in a Brazilian population. Pharmacogenetics and Genomics 2005;15:891–3. http://dx.doi.org/10.1097/01213011-200512000-00007, 16272961.
[60] Pliakas A.M., Carlson R.R., Neve R.L., Konradi C., Nestler E.J., Carlezon W.A. Jr.. Altered responsiveness to cocaine and increased immobility in the forced swim test associated with elevated cAMP response element-binding protein expression in nucleus accumbens. Journal of Neuroscience: The Official Journal of the Society for Neuroscience 2001;21:7397–403, 11549750.
[61] Tang W., Wesley M., Freeman W.M., Liang B., Hemby S.E. Alterations in ionotropic glutamate receptor subunits during binge cocaine self-administration and withdrawal in rats. Journal of Neurochemistry 2004;89:1021–33. http://dx.doi.org/10.1111/j.1471-4159.2004.02392.x, 15140200.
[62] Haile C.N., Hiroi N., Nestler E.J., Kosten T.A. Differential behavioral responses to cocaine are associated with dynamics of mesolimbic dopamine proteins in Lewis and Fischer 344 rats. Synapse (New York, N.Y.) 2001;41:179–90. http://dx.doi.org/10.1002/syn.1073, 11391778.
[63] Zhang D., Zhang L., Tang Y., Zhang Q., Lou D., Sharp F.R. et al. Repeated cocaine administration induces gene expression changes through the dopamine D1 receptors. Neuropsychopharmacology: Official Publication of the American College of Neuropsychopharmacology 2005;30:1443–54. http://dx.doi.org/10.1038/sj.npp.1300680, 15770241.
[64] Albertson D.N., Pruetz B., Schmidt C.J., Kuhn D.M., Kapatos G., Bannon M.J. Gene expression profile of the nucleus accumbens of human cocaine abusers: evidence for dysregulation of myelin. Journal of Neurochemistry 2004;88:1211–19. http://dx.doi.org/10.1046/j.1471-4159.2003.02247.x, 15009677.
[65] Bahi A., Dreyer J.L. Cocaine-induced expression changes of axon guidance molecules in the adult rat brain. Molecular and Cellular Neuroscience 2005;28:275–91.
[66] Yuferov V., Kroslak T., Laforge K.S., Zhou Y., Ho A., Kreek M.J. Differential gene expression in the rat caudate putamen after "binge" cocaine administration: advantage of triplicate microarray analysis. Synapse (New York, N.Y.) 2003;48:157–69. http://dx.doi.org/10.1002/syn.10198, 12687634.
[67] Yuferov V., Nielsen D., Butelman E., Kreek M.J. Microarray studies of psychostimulant-induced changes in gene expression. Addiction Biology 2005;10:101–18. http://dx.doi.org/10.1080/13556210412331308976, 15849024.
[68] Zhu W.L., Wang S.J., Liu M.M., Shi H.S., Zhang R.X., Liu J.F. et al. Glycine site N-methyl-D-aspartate receptor antagonist 7-CTKA produces rapid antidepressant-like effects in male rats. Journal of Psychiatry & Neuroscience: JPN 2013;38:306–16. http://dx.doi.org/10.1503/jpn.120228, 23611177.
[69] Cole A.R., Causeret F., Yadirgi G., Hastie C.J., McLauchlan H., McManus E.J. et al. Distinct priming kinases contribute to differential regulation of collapsin response mediator proteins by glycogen synthase kinase-3 in vivo. Journal of Biological Chemistry 2006;281, 16611631.
[70] Mash D.C., Adi N., Duque L., Pablo J., Kumar M., Ervin F.R. Alpha synuclein protein levels are increased in serum from recently abstinent cocaine abusers. Drug and Alcohol Dependence 2008;94:246–50, 18055133.
[71] Ryan B.J., Lourenco-Venda L., Crabtree M.J., Hale A.B., Channon K.M., Wade-Martins R. Alpha-synuclein and mitochondrial bioenergetics regulate tetrahydrobiopterin levels in a human dopaminergic model of PD. Free Radical Biology & Medicine 2013.
[72] Goumon Y., Muller A., Glattard E., Marban C., Gasnier C., Strub J.M. et al. Identification of morphine-6-glucuronide in chromaffin cell secretory granules. Journal of Biological Chemistry 2006;281:8082–9. http://dx.doi.org/10.1074/jbc.M502298200, 16434406.
[73] Duncan J.E., Lytle N.K., Zuniga A., Goldstein L.S. The microtubule regulatory protein stathmin is required to maintain the integrity of axonal microtubules in Drosophila. PLoS ONE 2013;8:e68324. http://dx.doi.org/10.1371/journal.pone.0068324, 23840848.
[74] Brand F., Schumacher S., Kant S., Menon M.B., Simon R., Turgeon B. et al. The extracellular signal-regulated kinase 3 (mitogen-activated protein kinase 6 [MAPK6])-MAPK-activated protein kinase 5 signaling complex regulates septin function and dendrite morphology. Molecular and Cellular Biology 2012;32:2467–78. http://dx.doi.org/10.1128/MCB.06633-11, 22508986.
[75] Itzhak Y. Role of the NMDA receptor and nitric oxide in memory reconsolidation of cocaine-induced conditioned place preference in mice. Annals of the New York Academy of Sciences 2008;1139:350–7. http://dx.doi.org/10.1196/annals.1432.051, 18991881.
[76] Addy N.A., Bahi A., Taylor J.R., Picciotto M.R. Administration of the calcineurin inhibitor cyclosporine modulates cocaine-induced locomotor activity in rats. Psychopharmacology 2008;200:129–39. http://dx.doi.org/10.1007/s00213-008-1189-5, 18587562.
[77] Lonart G., Simsek-Duran F. Deletion of synapsins I and II genes alters the size of vesicular pools and rabphilin phosphorylation. Brain Research 2006;1107:42–51. http://dx.doi.org/10.1016/j.brainres.2006.05.092, 16844103.
[78] Kauer J., Malenka R. Synaptic plasticity and addiction. Nature Reviews. Neuroscience 2007;8:844–58. http://dx.doi.org/10.1038/nrn2234, 17948030.
[79] Wong W., Scott J. AKAP signalling complexes: focal points in space and time. Nature Reviews. Molecular Cell Biology 2004;5:959–70. http://dx.doi.org/10.1038/nrm1527, 15573134.
[80] Mi Y.J., Hou B., Liao Q.M., Ma Y., Luo Q., Dai Y.K. et al. Amino-Nogo-A antagonizes reactive oxygen species generation and protects immature primary cortical neurons from oxidative toxicity. Cell Death and Differentiation 2012;19:1175–86. http://dx.doi.org/10.1038/cdd.2011.206, 22261619.
[81] Heizmann C. Calcium-binding proteins: basic concepts and clinical implications. General Physiology and Biophysics 1992;11:411–25, 1291444.
[82] Funakoshi-Hirose I., Aki T., Unuma K., Funakoshi T., Noritake K., Uemura K. Distinct effects of methamphetamine on autophagy-lysosome and ubiquitin-proteasome systems in HL-1 cultured mouse atrial cardiomyocytes. Toxicology 2013;312:74–82. http://dx.doi.org/10.1016/j.tox.2013.07.016, 23933405.
[83] Robinson T.E., Berridge K.C. The neural basis of drug craving: an incentive-sensitization theory of addiction. Brain Research. Brain Research Reviews 1993;18:247–91. http://dx.doi.org/10.1016/0165-0173(93)90013-P, 8401595.
[84] Krasnova I.N., Cadet J.L. Methamphetamine toxicity and messengers of death. Brain Research Reviews 2009;60:379–407. http://dx.doi.org/10.1016/j.brainresrev.2009.03.002, 19328213.
[85] Darke S., Kaye S., McKetin R., Duflou J. Major physical and psychological harms of methamphetamine use. Drug and Alcohol Review 2008;27:253–62. http://dx.doi.org/10.1080/09595230801923702, 18368606.
[86] Scott J., Woods S., Matt G., Meyer R., Heaton R., Atkinson J. et al. Neurocognitive effects of methamphetamine: a critical review and meta-analysis. Neuropsychology Review 2007;17:275–97. http://dx.doi.org/10.1007/s11065-007-9031-0, 17694436.
[87] Gonzalez R., Rippeth J.D., Carey C.L., Heaton R.K., Moore D.J., Schweinsburg B.C. et al. Neurocognitive performance of methamphetamine users discordant for history of marijuana exposure. Drug and Alcohol Dependence 2004;76:181–90. http://dx.doi.org/10.1016/j.drugalcdep.2004.04.014, 15488342.
[88] Matejovska I., Bernaskova K., Slamberova R. Effect of prenatal methamphetamine exposure and challenge dose of the same drug in adulthood on epileptiform activity induced by electrical stimulation in female rats. Neuroscience 2014;257:130–8. http://dx.doi.org/10.1016/j.neuroscience.2013.10.069, 24215975.
[89] Volkow N.D., Chang L., Wang G.-J., Fowler J.S., Leonido-Yee M., Franceschi D. et al. Association of dopamine transporter reduction with psychomotor impairment in methamphetamine abusers. American Journal of Psychiatry 2001;158:377–82, 11229977.
[90] Wilson J., Kalasinsky K., Levey A., Bergeron C., Reiber G., Anthony R. et al. Striatal dopamine nerve terminal markers in human, chronic methamphetamine users. Nature Medicine 1996;2:699–703. http://dx.doi.org/10.1038/nm0696-699, 8640565.
[91] Deng X., Cai N.-S., McCoy M.T., Chen W., Trush M.A., Cadet J.L. Methamphetamine induces apoptosis in an immortalized rat striatal cell line by activating the mitochondrial cell death pathway. Neuropharmacology 2002;42:837–45. http://dx.doi.org/10.1016/S0028-3908(02)00034-5, 12015210.
[92] Broening H.W., Vorhees C.V. Effect of methamphetamine on glutamate-positive neurons in the adult and developing rat somatosensory cortex. Synapse (New York, N.Y.) 1996;23:328–34. http://dx.doi.org/10.1002/(SICI)1098-2396(199608)23:4<328::AID-SYN11>3.0.CO;2-T, 8855518.
[93] Stumm G., Schlegel J., Schafer T., Wurz C., Mennel H.D., Krieg J.C. et al. Amphetamines induce apoptosis and regulation of bcl-x splice variants in neocortical neurons. FASEB Journal: Official Publication of the Federation of American Societies for Experimental Biology 1999;13:1065–72, 10336889.
[94] Sekine Y., Ouchi Y., Takei N., Yoshikawa E., Nakamura K., Futatsubashi M. et al. Brain serotonin transporter density and aggression in abstinent methamphetamine abusers. Archives of General Psychiatry 2006;63:90–100,

16389202.
[95] Ballmaier M., Sowell E.R., Thompson P.M., Kumar A., Narr K.L., Lavretsky H. et al. Mapping brain size and cortical gray matter changes in elderly depression. Biological Psychiatry 2004;55:382–9. http://dx.doi.org/10.1016/j.biopsych.2003.09.004, 14960291.
[96] Ernst T., Chang L., Leonido-Yee M., Speck O. Evidence for long-term neurotoxicity associated with methamphetamine abuse: a 1H MRS study. Neurology 2000;54:1344–9. http://dx.doi.org/10.1212/WNL.54.6.1344, 10746608.
[97] Sekine Y., Ouchi Y., Sugihara G., Takei N., Yoshikawa E., Nakamura K. et al. Methamphetamine causes microglial activation in the brains of human abusers. Journal of Neuroscience: The Official Journal of the Society for Neuroscience 2008;28:5756–61. http://dx.doi.org/10.1523/JNEUROSCI.1179-08.2008, 18509037.
[98] Monte D.A.D., Royland E., Jakowec M.W., Langston J.W. Role of nitric oxide in methamphetamine neurotoxicity: protection by 7-nitroindazole, an inhibitor of neuronal nitric oxide synthase. Journal of Neurochemistry 1996;67:2443–50, 8931477.
[99] Staszewski R.D., Yamamoto B.K. Methamphetamine-induced spectrin proteolysis in the rat striatum. Journal of Neurochemistry 2006;96:1267–76. http://dx.doi.org/10.1111/j.1471-4159.2005.03618.x, 16417574.
[100] Jayanthi S., Deng X., Noailles P.A., Ladenheim B., Cadet J.L. Methamphetamine induces neuronal apoptosis via cross-talks between endoplasmic reticulum and mitochondria-dependent death cascades. FASEB Journal: Official Publication of the Federation of American Societies for Experimental Biology 2004;18:238–51. http://dx.doi.org/10.1096/fj.03-0295com, 14769818.
[101] Jayanthi S., Deng X., Bordelon M., Mccoy M.T., Cadet J.L. Methamphetamine causes differential regulation of pro-death and anti-death Bcl-2 genes in the mouse neocortex. FASEB Journal 2001;15:1745–52. http://dx.doi.org/10.1096/fj.01-0025com.
[102] Poon H.F., Shepherd H.M., Reed T.T., Calabrese V., Stella A.M.G., Pennisi G. et al. Proteomics analysis provides insight into caloric restriction mediated oxidation and expression of brain proteins associated with age-related impaired cellular processes: mitochondrial dysfunction, glutamate dysregulation and impaired protein synthesis. Neurobiology of Aging 2006;27:1020–34. http://dx.doi.org/10.1016/j.neurobiolaging.2005.05.014, 15996793.
[103] Lawen A. Apoptosis – an introduction. BioEssays: News and Reviews in Molecular, Cellular and Developmental Biology 2003;25:888–96. http://dx.doi.org/10.1002/bies.10329, 12938178.
[104] Harada T., Harada C., Wang Y.L., Osaka H., Amanai K., Tanaka K. et al. Role of ubiquitin carboxy terminal hydrolase-L1 in neural cell apoptosis induced by ischemic retinal injury in vivo. American Journal of Pathology 2004;164:59–64. http://dx.doi.org/10.1016/S0002-9440(10)63096-9, 14695319.
[105] Choi J., Levey A.I., Weintraub S.T., Rees H.D., Gearing M., Chin L.S. et al. Oxidative modifications and down-regulation of ubiquitin carboxyl-terminal hydrolase L1 associated with idiopathic Parkinson's and Alzheimer's diseases. Journal of Biological Chemistry 2004;279:13256–64, 14722078.
[106] Fornai F.L., Gesi M., Ferrucci M., Lazzeri G., Capobianco L., de Blasi A. et al. Similarities between methamphetamine toxicity and proteasome inhibition. Annals of the New York Academy of Sciences 2004;1025:162–70. http://dx.doi.org/10.1196/annals.1316.021, 15542714.
[107] Messa M., Congia S., Defranchi E., Valtorta F., Fassio A., Onofri F. et al. Tyrosine phosphorylation of synapsin I by Src regulates synaptic-vesicle trafficking. Journal of Cell Science 2010;123:2256–65. http://dx.doi.org/10.1242/jcs.068445, 20530578.
[108] Coleman W., Bill C., Simsek-Duran F., Lonart G., Samigullin D., Bykhovskaia M. Synapsin II and calcium regulate vesicle docking and the cross-talk between vesicle pools at the mouse motor terminals. Journal of Physiology 2008;586:4649–73. http://dx.doi.org/10.1113/jphysiol.2008.154666, 18669537.
[109] Lindholm D., Mercer E.A., Yu L.Y., Chen Y., Kukkonen J., Korhonen L. et al. Neuronal apoptosis inhibitory protein: structural requirements for hippocalcin binding and effects on survival of NGF-dependent sympathetic neurons. Biochimica et Biophysica Acta 2002;1600:138–47. http://dx.doi.org/10.1016/S1570-9639(02)00454-5, 12445469.
[110] Liao P.C., Kuo Y.M., Hsu H.C., Cherng C.G., Yu L. Local proteins associated with methamphetamine-induced nigrostriatal dopaminergic neurotoxicity. Journal of Neurochemistry 2005;95:160–8. http://dx.doi.org/10.1111/j.1471-4159.2005.03346.x, 16181420.
[111] Machida Y., Chiba T., Takayanagi A., Tanaka Y., Asanuma M., Ogawa N. et al. Common anti-apoptotic roles of Parkin and alpha-synuclein in human dopaminergic cells. Biochemical and Biophysical Research Communications 2005;24:233–40, 15896322.
[112] Rakhit R.C. Structure, folding, and misfolding of Cu,Zn superoxide dismutase in amyotrophic lateral sclerosis. Biochimica et Biophysica Acta 2006;1762:1025–37. http://dx.doi.org/10.1016/j.bbadis.2006.05.004, 16814528.
[113] Zhu J., Xu W., Wang J., Ali S.F., Angulo J.A. The neurokinin-1 receptor modulates the methamphetamine-induced striatal apoptosis and nitric oxide formation in mice. Journal of Neurochemistry 2009;111:656–68. http://dx.doi.org/10.1111/j.1471-4159.2009.06330.x, 19682209.
[114] Schmidt G., Wittinghofer A. Priming of PC12 cells for semiquantitative microinjection studies involving Ras. FEBS Letters 2000;474:184–8. http://dx.doi.org/10.1016/S0014-5793(00)01602-1, 10838082.
[115] Sun K.H., Chang K.H., Clawson S., Ghosh S., Mirzaei H., Regnier F. et al. Glutathione-S-transferase P1 is a critical regulator of Cdk5 kinase activity. Journal of Neurochemistry 2011;118:902–14. http://dx.doi.org/10.1111/j.1471-4159.2011.07343.x, 21668448.
[116] Nath R., Raser K.J., Stafford D., Hajimohammadreza I., Posner A., Allen H. et al. Non-erythroid α-spectrin breakdown by calpain and interleukin 1β-converting-enzyme-like protease(s) in apoptotic cells: contributory roles of both protease families in neuronal apoptosis. Biochemical Journal 1996;319:683–90, 8920967.
[117] Wallace T.L., Vorhees C.V., Zemlan F.P., Gudelsky G.A. Methamphetamine enhances the cleavage of the cytoskeletal protein tau in the rat brain. Neuroscience 2003;116:1063–8. http://dx.doi.org/10.1016/S0306-4522(02)00795-9, 12617947.
[118] Shacka J.J.L., Xie Z.L., Uchiyama Y., Roth K.A., Zhang J. Kainic acid induces early and transient autophagic stress in mouse hippocampus. Neuroscience Letters 2007;414:57–60. http://dx.doi.org/10.1016/j.neulet.2006.12.025, 17223264.
[119] Kanthasamy A.A., Ali S.F., Kanthasamy A.G. Methamphetamine induces autophagy and apoptosis in a mes- encephalic dopaminergic neuronal culture model: role of cathepsin-D in methamphetamine-induced apoptotic cell death. Annals of the New York Academy of Sciences 2006;1074:234–44. http://dx.doi.org/10.1196/annals.1369.022, 17105920.
[120] Chen Q.W., Thompson S.N., Hall E.D., Guttmann R.P. Identification and characterization of PEBP as a calpain substrate. Journal of Neurochemistry 2006;99:1133–41. http://dx.doi.org/10.1111/j.1471-4159.2006.04160.x, 17018026.
[121] Zhang Z.O., Shankar S., Kobeissy F., Tei F., Hayes R.L., Wang K.K. Calpain-mediated collapsin response mediator protein-1, -2, and -4 proteolysis after neurotoxic and traumatic brain injury. Journal of Neurotrauma 2007;24:460–72. http://dx.doi.org/10.1089/neu.2006.0078, 17402852.
[122] Aitken A. 14-3-3 proteins on the MAP. Trends in Biochemical Sciences 1995;20:95–7.
[123] Narita M., Akai H., Nagumo Y., Sunagawa N., Hasebe K., Nagase H. et al. Implications of protein kinase C in the nucleus accumbens in the development of sensitization to methamphetamine in rats. Neuroscience 2004;127:941–8. http://dx.doi.org/10.1016/j.neuroscience.2004.06.017, 15312906.
[124] Tsukamoto T., Iyo M., Tani K., Sekine Y., Hashimoto K., Ohashi Y. et al. The effects of FK506, a specific calcineurin inhibitor, on methamphetamine-induced behavioral change and its sensitization in rats. Psychopharmacology 2001;158:107–13. http://dx.doi.org/10.1007/s002130100870, 11702083.
[125] Borlongan C., Freeman T., Scorcia T., Sherman K., Olanow W., Cahill D. et al. Cyclosporine-A increases spontaneous and dopamine agonist-induced locomotor behavior in normal rats. Cell Transplantation 1995;4:65–73. http://dx.doi.org/10.1016/0963-6897(94)00040-Q, 7728335.
[126] Zeng K., Wang X., Wan Y., Yan Y. Enhanced synaptic vesicle traffic in hippocampus of phenytoin-resistant kindled rats. Neurochemical Research 2009;34:899–904. http://dx.doi.org/10.1007/s11064-008-9856-9, 18841467.
[127] Janz R., Goda Y., Geppert M., Missler M., Sudhof T.C. SV2A and SV2B function as redundant Ca2+ regulators in neurotransmitter release. Neuron 1999;24:1003–16. http://dx.doi.org/10.1016/S0896-6273(00)81046-6, 10624962.
[128] Hanaya R., Hosoyama H., Sugata S., Tokudome M., Hirano H., Tokimura H. et al. Low distribution of synaptic vesicle protein 2A and synaptotagimin-1 in the cerebral cortex and hippocampus of spontaneously epileptic rats exhibiting both tonic convulsion and absence seizure. Neuroscience 2012;221:12–20. http://dx.doi.org/10.1016/j.neuroscience.2012.06.058, 22766234.
[129] Bignone P., King M., Pinder J., Baines A. Phosphorylation of a threonine unique to the short C-terminal isoform of betaII-spectrin links regulation of alpha-beta spectrin interaction to neuritogenesis. Journal of Biological Chemistry 2007;282:888–96. http://dx.doi.org/10.1074/jbc.M605920200, 17088250.
[130] Voas M., Lyons D., Naylor S., Arana N., Rasband M., Talbot W. alphaII-spectrin is essential for assembly of the nodes of Ranvier in myelinated axons. Current Biology 2007;17:562–8. http://dx.doi.org/10.1016/j.cub.2007.01.071, 17331725.
[131] Selzer E., Wilfing A., Schiferer A., Hermann M., Grubeck-LoeBestein B., Freissmuth M. Stimulation of human thyroid growth via the inhibitory guanine nucleotide binding (G) protein G_i: constitutive expression of the G-protein α subunit $G_{i\alpha\text{-}i}$ in autonomous adenoma. Proceedings of the National Academy of Sciences 1993;90:1609–13. http://dx.doi.org/10.1073/pnas.90.4.1609.
[132] Demetriou M., Binkert C., Sukhu B., Tenenbaum H.C., Dennis J.W. Fetuin/alpha2-HS glycoprotein is a transforming growth factor-beta type II receptor mimic and cytokine antagonist. Journal of Biological Chemistry 1996;271:12755–61. http://dx.doi.org/10.1074/jbc.271.22.12755, 8662721.
[133] Rasul S., Wagner L., Kautzky-Willer A. Fetuin-A and angiopoietins in obesity and type 2 diabetes mellitus. Endocrine 2012;42:496–505. http://dx.doi.org/10.1007/s12020-012-9754-4, 22820893.
[134] Way G., Morrice N., Smythe C., O'Sullivan A.J. Purification and identification of secernin, a novel cytosolic protein that regulates exocytosis in mast cells. Molecular Biology of the Cell 2002;13:3344–54. http://dx.doi.org/10.1091/mbc.E01-10-0094, 12221138.
[135] Suda T., Tsunoda T., Uchida N., Watanabe T., Hasegawa S., Satoh S. et al. Identification of secernin 1 as a novel immunotherapy target for gastric cancer using the expression profiles of cDNA microarray. Cancer Science 2006;97:411–19. http://dx.doi.org/10.1111/j.1349-7006.2006.00194.x, 16630140.
[136] Suehara Y., Tochigi N., Kubota D., Kikuta K., Nakayama R., Seki K. et al. Secernin-1 as a novel prognostic biomarker candidate of synovial sarcoma revealed by proteomics. Journal of Proteomics 2011;74:829–42. http://dx.doi.org/10.1016/j.jprot.2011.02.033, 21385630.
[137] Landry M. MDMA: a review of epidemiologic data. Journal of Psychoactive Drugs 2002;34:163–9. http://dx.doi.org/10.1080/02791072.2002.10399950, 12691206.
[138] Reid L., Elifson K., Sterk C. Ecstasy and gateway drugs: initiating the use of

ecstasy and other drugs. Annals of Epidemiology 2007;17:74–80. http://dx.doi.org/10.1016/j.annepidem.2006.07.013, 17140814.
[139] Schifano F., Corkery J., Deluca P., Oyefeso A., Ghodse A. Ecstasy (MDMA, MDA, MDEA, MBDB) consumption, seizures, related offences, prices, dosage levels and deaths in the UK (1994–2003). Journal of Psychopharmacology (Oxford, England) 2006;20:456–63, 16574720.
[140] Ellis A., Wendon J., Portmann B., Williams R. Acute liver damage and ecstasy ingestion. Gut 1996;38:454–8. http://dx.doi.org/10.1136/gut.38.3.454, 8675102.
[141] Green A., Mechan A., Elliott J., Shea E., Colado M. The pharmacology and clinical pharmacology of 3,4-methylenedioxymethamphetamine (MDMA, "Ecstasy"). Pharmacological Reviews 2003;55:463–508. http://dx.doi.org/10.1124/pr.55.3.3, 12869661.
[142] Gramage E., Rossi L., Granado N., Moratalla R., Herradón G. Genetic inactivation of pleiotrophin triggers amphetamine- induced cell loss in the substantia nigra and enhances amphetamine neurotoxicity in the striatum. Neuroscience 2010;170:308–16. http://dx.doi.org/10.1016/j.neuroscience.2010.06.078, 20620199.
[143] Cadet J., Krasnova I., Jayanthi S., Lyles J. Neurotoxicity of substituted amphetamines: molecular and cellular mechanisms. Neurotoxicity Research 2007;11:183–202. http://dx.doi.org/10.1007/BF03033567, 17449459.
[144] Hansen J., Riddle E., Sandoval V., Brown J., Gibb J., Hanson G. et al. Methylenedioxymethamphetamine decreases plasmalemmal and vesicular dopamine transport: mechanisms and implications for neurotoxicity. Journal of Pharmacology and Experimental Therapeutics 2002;300:1093–100. http://dx.doi.org/10.1124/jpet.300.3.1093, 11861820.
[145] Venugopal S., Chen J., Zhang Y., Clemens D., Follenzi A., Zern M. Role of MAPK phosphatase-1 in sustained activation of JNK during ethanol-induced apoptosis in hepatocyte-like VL-17A cells. Journal of Biological Chemistry 2007;282:31900–8. http://dx.doi.org/10.1074/jbc.M703729200, 17848570.
[146] Widmer R., Kaiser B., Engels M., Jung T., Grune T. Hyperammonemia causes protein oxidation and enhanced proteasomal activity in response to mitochondria-mediated oxidative stress in rat primary astrocytes. Archives of Biochemistry and Biophysics 2007;464:1–11. http://dx.doi.org/10.1016/j.abb.2007.03.027, 17475207.
[147] Berlett B., Stadtman E. Protein oxidation in aging, disease, and oxidative stress. Journal of Biological Chemistry 1997;272:20313–16. http://dx.doi.org/10.1074/jbc.272.33.20313, 9252331.
[148] Paris J., Cunningham K. Lack of serotonin neurotoxicity after intraraphe microinjection of (+)-3,4-methylenedioxymethamphetamine (MDMA). Brain Research Bulletin 1992;28:115–19. http://dx.doi.org/10.1016/0361-9230(92)90237-R, 1347247.
[149] Schmidt C., Taylor V. Direct central effects of acute methylenedioxymethamphetamine on serotonergic neurons. European Journal of Pharmacology 1988;156:121–31. http://dx.doi.org/10.1016/0014-2999(88)90154-9, 2463176.
[150] Alves E., Summavielle T., Alves C., Gomes-da-Silva J., Barata J., Fernandes E. et al. Monoamine oxidase-B mediates ecstasy-induced neurotoxic effects to adolescent rat brain mitochondria. Journal of Neuroscience: The Official Journal of the Society for Neuroscience 2007;27:10203–10. http://dx.doi.org/10.1523/JNEUROSCI.2645-07.2007, 17881526.
[151] Fisher A., Labenski M., Malladi S., Gokhale V., Bowen M., Milleron R. et al. Quinone electrophiles selectively adduct "electrophile binding motifs" within cytochrome c. Biochemistry 2007;46:11090–100, 17824617.
[152] Montiel-Duarte C., Ansorena E., Lopez-Zabalza M., Cenarruzabeitia E., Iraburu M. Role of reactive oxygen species, glutathione and NF-kappaB in apoptosis induced by 3,4-methylenedioxy methamphetamine ("Ecstasy") on hepatic stellate cells. Biochemical Pharmacology 2004;67:1025–33. http://dx.doi.org/10.1016/j.bcp.2003.10.020, 15006539.
[153] Pacher P., Beckman J., Liaudet L. Nitric oxide and peroxynitrite in health and disease. Physiological Reviews 2007;87:315–424. http://dx.doi.org/10.1152/physrev.00029.2006, 17237348.
[154] Oesterheld J., Armstrong S., Cozza K. Ecstasy: pharmacodynamic and pharmacokinetic interactions. Psychosomatics 2004;45:84–7. http://dx.doi.org/10.1176/appi.psy.45.1.84, 14709765.
[155] Jaim-Etcheverry G., Zieher L. DSP-4: a novel compound with neurotoxic effects on noradrenergic neurons of adult and developing rats. Brain Research 1980;188:513–23. http://dx.doi.org/10.1016/0006-8993(80)90049-9, 7370771.
[156] Kuwayama K., Tsujikawa K., Miyaguchi H., Kanamori T., Iwata Y.T., Inoue H. Distribution measurements of 3,4-methylenedioxymethamphetamine and its metabolites in organs by matrix-assisted laser desorption/ionization imaging mass spectrometry using an automatic matrix spraying system with an air brush and a turntable. Analytical and Bioanalytical Chemistry 2012;404:1823–30. http://dx.doi.org/10.1007/s00216-012-6279-x, 22865009.
[157] da Silva D.D., Silva E., Carvalho F., Carmo H. Mixtures of 3,4-methylenedioxymethamphetamine (Ecstasy) and its major human metabolites act additively to induce significant toxicity to liver cells when combined at low, non-cytotoxic concentrations. Journal of Applied Toxicology: JAT 2014;34:618–27. http://dx.doi.org/10.1002/jat.2885, 23670916.
[158] Martins J., Castelo-Branco M., Batista A., Oliveiros B., Santiago A., Galvão J. et al. Effects of 3,4-methylenedioxymethamphetamine administration on retinal physiology in the rat. PLoS ONE 2011;6, 22216322.
[159] Ninković M., Selaković V., Dukić M., Milosavljević P., Vasiljević I., Jovanović M. et al. Oxidative stress in rat kidneys due to 3,4-methylenedioxymetamphetamine (ecstasy) toxicity. Nephrology (Carlton, Vic.) 2008;13:33–7, 18199099.
[160] Substance Abuse and Mental Health Services Administration, Results from the 2012 National Survey on Drug Use and Health: Summary of National Findings, NSDUH Series H-46, HHS Publication No. (SMA) 13-4795. Rockville, MD: Substance Abuse and Mental Health Services Administration, 2013.
[161] Hemby, S.E., 2006. Assessment of genome and proteome profiles in cocaine abuse. 158, 173-195.