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The Role of Secretory Phospholipase A_2 in the Central Nervous System and Neurological Diseases

Tatsurou Yagami · Yasuhiro Yamamoto · Hiromi Koma

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Abstract Secretory phospholipase A_2 ($sPLA_2$s) are small secreted proteins (14–18 kDa) and require submillimolar levels of Ca^{2+} for liberating arachidonic acid from cell membrane lipids. In addition to the enzymatic function, $sPLA_2$ can exert various biological responses by binding to specific receptors. Physiologically, $sPLA_2$s play important roles on the neurotransmission in the central nervous system and the neuritogenesis in the peripheral nervous system. Pathologically, $sPLA_2$s are involved in the neurodegenerative diseases (e.g., Alzheimer's disease) and cerebrovascular diseases (e.g., stoke). The common pathology (e.g., neuronal apoptosis) of Alzheimer's disease and stroke coexists in the mixed dementia, suggesting common pathogenic mechanisms of the two neurological diseases. Among mammalian $sPLA_2$s, $sPLA_2$-IB and $sPLA_2$-IIA induce neuronal apoptosis in rat cortical neurons. The excess influx of calcium into neurons via L-type voltage-dependent Ca^{2+} channels mediates the two $sPLA_2$-induced apoptosis. The elevated concentration of intracellular calcium activates PKC, MAPK and cytosolic PLA_2. Moreover, it is linked with the production of reactive oxygen species and apoptosis through activation of the superoxide producing enzyme NADPH oxidase. NADPH oxidase is involved in the neurotoxicity of amyloid β peptide, which impairs synaptic plasticity long before its deposition in the form of amyloid plaques of Alzheimer's disease. In turn, reactive oxygen species from NADPH oxidase can stimulate ERK1/2 phosphorylation and activation of $cPLA_2$ and result in a release of arachidonic acid. $sPLA_2$ is up-regulated in both Alzheimer's disease and cerebrovascular disease, suggesting the involvement of $sPLA_2$ in the common pathogenic mechanisms of the two diseases. Thus, our review presents evidences for pathophysiological roles of $sPLA_2$ in the central nervous system and neurological diseases.

Keywords Alzheimer's disease · Cerebrovascular disease · Long-term potentiation · L-type voltage-dependent Ca^{2+} channels · Neuronal apoptosis · Phospholipase A_2

Abbreviations

AA	Arachidonic acid
AD	Alzheimer's disease
AMPA	Amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid
APP	Amyloid precursor protein
CaMK II	Calcium/calmodulin-dependent protein kinase II
COX	Cyclooxygenase
CVD	Cerebrovascular disease
$iPLA_2$	Ca^{2+}-independent cytosolic phospholipase A_2
ER	Endoplasmic reticulum
FAβ	Fibrillar Aβ
$[Ca^{2+}]_i$	Intracellular Ca^{2+} concentration
IL-1β	Interleukin-1β
LGCC	Ligand-gated calcium channel
LTP	Long-term potentiation
L-VDCC	L-Type voltage-dependent Ca^{2+} channels
MAPK	Mitogen-activated protein kinases
NMDA	*N*-Methyl-D-aspartate
NFTs	Neurofibrillary tangles
PAF	Platelet activating factor
PLA_2	Phospholipase A_2
PG	Prostaglandin
ROS	Reactive oxygen species
$sPLA_2$	Secreted phospholipase A_2
$sPLA_2R$	Secretory phospholipase A_2 receptor

T. Yagami (✉) · Y. Yamamoto · H. Koma
Division of Physiology, Faculty of Pharmaceutical Sciences, Himeji Dokkyo University, 2-1-7, Kami-ohno, Himeji, Hyogo 670-8524, Japan
e-mail: yagami@himeji-du.ac.jp

Background

Phospholipase A_2 (PLA_2) belongs to a family of enzymes that catalyze the cleavage of fatty acids from the sn-2 position of glycerophospholipids to produce free fatty acids and lysophospholipids. PLA_2s participate in a wide variety of physiological processes, including phospholipid digestion, remodeling of cell membranes and host defense. According to their biochemical features such as cellular localization, requirement of Ca^{2+}, substrate specificity and the primary structure, more than 19 different isoforms of mammalian PLA_2 are classified into several families, including low molecular weight secretory PLA_2 ($sPLA_2$), Ca^{2+}-sensitive arachidonoyl-specific 85-kDa cytosolic PLA_2 ($cPLA_2$), Ca^{2+}-independent PLA_2 ($iPLA_2$), plasmalogen-selective PLA_2 and platelet-activating factor-acetylhydrolase [1]. The common structural and catalytic properties of mammalian $sPLA_2$s are: the presence of 6–8 disulfide bonds, highly conserved catalytical dyad His–Asp and "Ca binding loop", a requirement for millimolar concentration of Ca^{2+} for catalysi, and a broad specificity for phospholipids with different polar head groups and fatty acid chains. In addition to the digestive function, $sPLA_2$ can exert various biological responses through the binding to the cell surface PLA_2 receptor [2]. In the mammalian nervous system, nine $sPLA_2$ (IB, IIA, IIC, IIE, IIF, III, V, X and XII), $cPLA_2$ (IV), $iPLA_2$ (VI) and platelet-activating factor (PAF)-acetylhydrolase (VII, VIII) have been detected.

Occurrence of $sPLA_2$s in Mammalian Nervous Tissues and Neural Cells

The $sPLA_2$s are small secreted proteins (14–18 kDa) and use an active site histidine and absolutely require submillimolar levels of Ca^{2+} for catalysis. In close proximity to the catalytic histidine, there is a conserved aspartate. The two amino acids form a His/Asp dyad. Mammalian PLA_2s do not show a distinct preference for particular fatty acids, whereas there is some specificity for certain head groups of the phospholipid substrate: generally, $sPLA_2$s show a high activity with anionic phospholipid but only the $sPLA_2$-V and $sPLA_2$-X PLA_2s also hydrolyze phosphatidylcholine vesicles.

$sPLA_2$-IB

The human $sPLA_2$-IB consists of 148 amino acids (calculated molecular mass [M_r]=16,800). The gene of $sPLA_2$-IB maps to chromosome 12q23-24. $sPLA_2$s are Ca^{2+}-dependent for catalytic activity (generally requiring millimolar Ca^{2+} concentrations) [3], consistent with their extracellular activity and millimolar Ca^{2+} concentrations in the extracellular space [4]. Evidence also points to an intracellular functional role $sPLA_2$ in caveolae-containing compartments around the nucleus of immune cells and in the nucleus of astrocytes and neurons [5]. $sPLA_2$-IB has a unique five amino acid extension termed the pancreatic loop in the middle part of the molecule and a group I-specific disulfide between Cys^{11} and Cys^{77}. $sPLA_2$-IB is produced as an inactive pro-enzyme (pro-$sPLA_2$-IB) and is activated by proteolytic enzymes such as trypsin and plasmin. Given its abundance in digestive organs, the major physiological function of $sPLA_2$-IB has been thought to be the digestion of glycerophospholipids in nutrients [6]. Although $sPLA_2$-IB is originally purified and sequenced from pancreas or pancreatic juice of various mammals, it is also found in the brain [7]. Its mRNA is most abundant in the cerebral cortex, followed by the temporal lobe (Table 1). $sPLA_2$-IB is expressed in the neuronal cell body of cortex, hippocampus and cerebellum. Although $sPLA_2$-IB is not detected in the outer layer of cortex, it is abundant in layers 2–4. In the hippocampus, $sPLA_2$-IB is present in all CA regions as well as in the dentate gyrus [7].

$sPLA_2$-IIA

The human $sPLA_2$-IIA consists of 144 amino acids (M_r=16,083) and is most similar to $sPLA_2$-IB with respect to the number and positions of cysteine residues as well as overall identity (51 %) [8]. The gene of human $sPLA_2$-IIA maps to chromosome 1p35. Its enzymatic properties are almost identical to those of $sPLA_2$-IB in terms of Ca^{2+} requirement, optimal pH, substrate specificity, as well as high susceptibility to the $sPLA_2$ inhibitor indoxam. $sPLA_2$-IIA mRNA is detected in all brain region (Table 1). It is highly expressed in the brainstem and midbrain, and lowly in cerebellum and corpus striatum [9].

$sPLA_2$-IIC

The gene of human $sPLA_2$-IIC maps to chromosome 1p35.12. $sPLA_2$-IIC mRNA is exclusively expressed in the brain. As shown in Table 1, it is highly expressed in hypothalamus, cerebral cortex, midbrain, striatum, and hippocampus and lowly expressed in brainstem, thalamus and cerebellum [9]. However, protein of $sPLA_2$-IIC has not yet been detected and its function not also detected. The gene encoding for $sPLA_2$-IIC appears to be a pseudogene.

$sPLA_2$-IIE

The human $sPLA_2$-IIE consists of 123 amino acids (M_r=14,000) and is most similar to $sPLA_2$-IIA with respect to the number and positions of cysteine residues as well as overall identity (51 %) [8]. Its enzymatic properties are almost identical to those of $sPLA_2$-IIA in terms of Ca^{2+} requirement, optimal pH, substrate specificity, as well as high susceptibility to the $sPLA_2$ inhibitor indoxam. $sPLA_2$-IIE retains considerable catalytic activity at pH 5, with activity falling as the pH is

Table 1 $sPLA_2$s in nervous tissues and in neuronal cells

Tissues	IB [7]	IIA [9]	IIC [9]	IIE [11]	IIF [12]	III [14]	V [11]	X [11]	XII [19]
Brain	PCR			PCR	NB		PCR	PCR	NB
							ISH	ISH	
Neuron	PCR			PCR		WB	PCR	PCR	
						IH	PCR	ISH	
								IH	
Astrocyte	PCR						PCR		
							ISH		
Cerebral cortex	ISH		PCR	ISH		WB	PCR		
	IH					IH	ISH		
							IH		
Hippocampus	ISH			ISH			PCR	PCR	
							ISH	ISH	
							IH	IH	
Cerebellum	ISH	PCR	PCR	ISH		WB	PCR		
	IH	(Low)	(Low)			IH	ISH		
Striatum		PCR	PCR						
Thalamus			PCR						
			(Low)						
Hypothalamus			PCR						
Midbrain		PCR	PCR				PCR		
							ISH		
Brainstem		PCR	PCR						
		(Low)	(Low)						
Spinal cord						WB			
						IH			
DRG						WB			
						IH			

mRNA of $sPLA_2$ is detected by real time-polymerase chain reaction (RT-PCR), Northern blotting (NB) and in situ hybridization (ISH). Protein of $sPLA_2$ is detected by Western blotting (WB) and immunohistochemistry (IH)

raised above 6, suggesting its function in weakly acidic cellular compartments. No catalytic activity is detected for mouse $sPLA_2$-IIE in the absence of calcium [10]. The gene of human $sPLA_2$-IIE maps to chromosome 1p36.13. PLA_2-IIE mRNA is detected by RT-PCR in rat brain as well as in primary neuronal cultures [11]. In situ hybridization is carried out to identify the location of the $sPLA_2$ in rat brain. The mRNA expression of $sPLA_2$-IIE is found in cortex, hippocampus and cerebellum (Table 1).

$sPLA_2$-IIF

The full-length cDNA codes for a signal peptide of 20 amino acids followed by a mature protein of 148 amino acids containing all of the structural features of catalytically active $sPLA_2$-IIs [12]. The gene of human $sPLA_2$-IIF maps to chromosome 1p35. Its unique feature is the presence of a C-terminal extension of 23 amino acids containing a single cysteine. Human $sPLA_2$-IIF has been detected in patients with rheumatoid arthritis, but not in the brain. Mouse $sPLA_2$-IIF is strongly expressed during embryogenesis and in adult testis. In the mouse brain, Northern blot analysis reveals a very low level of $sPLA_2$-IIF [10]. $sPLA_2$-IIF retains considerable catalytic activity at pH 5, with activity falling as the pH is raised above 6, suggesting its function in weakly acidic cellular compartments. No catalytic activity is detected for mouse $sPLA_2$-IIF in the absence of calcium [10].

$sPLA_2$-III

Human $sPLA_2$-III homologous to the bee venom group III is identified by a systematic homology search in nucleic data bases [13]. The $sPLA_2$ domain is 31 % identical to bee venom $sPLA_2$ and displays all features of group III $sPLA_2$s including ten cysteines. The gene of $sPLA_2$-III maps to chromosome 22q. Its mRNA is highly expressed in kidney, heart, liver and skeletal muscle, but only weakly in the brain. $sPLA_2$-III is expressed in neuronal cells, such as peripheral neuronal fibers,

spinal dorsal root ganglia neurons and cerebellar Purkinje cells [14]. $sPLA_2$-III protein is highly expressed in the brainstem, spinal cord and cerebral cortex. $sPLA_2$-III protein is present in the "light membrane/cytosol" fraction, but not the nucleus, synaptosomal membrane or synaptic vesicle-enriched fractions. $sPLA_2$-III is immunolocalized to neurons in the cerebral cortex, Purkinje neurons in the cerebellar cortex, periaqueductal gray, red nucleus, spinal trigeminal nucleus and dorsal horn of the spinal cord. Electron microscopy of the spinal cord and cerebral cortex showed that $sPLA_2$-III is localized in dendrites or dendritic spines, that formed asymmetrical synapses with unlabeled, putatively glutamatergic, axon terminals.

$sPLA_2$-V

Human $sPLA_2$-V shows a high level of identity with $sPLA_2$-IIA, and its gene is located in chromosome 1 close to the gene of $sPLA_2$-IIA [15]. However, $sPLA_2$-V lacks one of the seven disulfide bridges found in similar $sPLA_2$ and, therefore, represents a class of enzymes distinct from $sPLA_2$-IIA. Cloned human $sPLA_2$-V has a signal peptide of 20 amino acids, and the matured protein (M_r=13,692) consists of 118 residues. $sPLA_2$-V effectively hydrolyzes phosphatidylcholine vesicles and the outer plasma membrane of mammalian cells. The gene of $sPLA_2$-V maps to chromosome 1p36–p34. Rat $sPLA_2$-V mRNA is detected in all brain regions, and highly expressed in the hippocampus [9]. Its protein is also found in the neurons in cerebral cortex and dentate gyrus [11], and in the Bergman glial cells in cerebellum [16]. Immunofluorescence analysis of isolated rat brain nuclei reveals that the enzyme is mainly localized in the nucleoplasm [17].

$sPLA_2$-X

Human $sPLA_2$-X shows a high level of identity with, and its gene is located in chromosome 16p13.1–p12 [18]. $sPLA_2$-X contains unique structures: (1) a long prepropeptide ending with an arginine doublet, (2) 16 cysteines located at positions that are characteristic of both $sPLA_2$-I and II, (3) a C-terminal extension typical of $sPLA_2$-IIs, and (4) the absence of elapid and pancreatic loops that are characteristic of $sPLA_2$-Is. Cloned human $sPLA_2$-X has a matured protein (calculated molecular mass of 13.6 kDa) consists of 123 residues. Contrary to the other $sPLA_2$, human $sPLA_2$-X is very acidic (pI=5.3) and possesses disulfide bridges typical to $sPLA_2$-IB and $sPLA_2$-IIA. Substrate preferences of $sPLA_2$-X are phosphatidylethanolamine and phosphatidylcholine liposomes rather than phosphatidylserine. $sPLA_2$-X is detected in neurons in cerebral cortex and dentate gyrus (Table 1). The level of $sPLA_2$-X is lower than those of other neuronal $sPLA_2$s in the central nervous system [11]. On the other hand, $sPLA_2$-X is expressed in the peripheral neuronal fibers and in the primary culture of dorsal root ganglia neurons.

$sPLA_2$-XII

The human $sPLA_2$-XII cDNA codes for a signal peptide of 22 amino acids followed by a mature protein of 167 amino acids that displays a poor sequence homology with other known $sPLA_2$s apart from the active site region [19]. The gene of human $sPLA_2$-XII maps to chromosome 4q25. The Northern blot analysis of tissue distribution of human $sPLA_2$-XII reveals a major transcript in heart, skeletal muscle, and kidney and lower levels in other tissues including the brain.

Physiological Roles of Secretory Phospholipase A_2 in the Central Nervous System

Neuritogenesis

The enrichment of cytoplasmic PLA_2 in growth cones suggests its plausible involvement in the regulation of growth cone function [20]. Rat pheochromocytoma PC12 cells are often used for the detailed analysis of neuronal functions (Table 2). They are differentiated into adherent cells similar to sympathetic neurons in response to nerve growth factor. Nerve growth factor induces neurite outgrowth and extends processes of PC 12 cells similar to neurites. Adenoviral expression of human $sPLA_2$-III in PC12 cells or dorsal root ganglia explants facilitates neurite-like outgrowth, whereas expression of a catalytically inactive $sPLA_2$-III mutant or use of $sPLA_2$-III-directed small interfering RNA (siRNA) reduces nerve growth factor-induced the extension of process [14]. Mammalian $sPLA_2$-V and -X also exhibit this effect in PC12 cells [21], and the extension is correlated to its activity rather than to a receptor-mediated mechanism [21]. The neuritogenic activity of $sPLA_2$ is mediated by generation of lysophosphatidylcholine and subsequent activation of G2A [22]. A G-protein-coupled receptor G2A mediates the signaling of lysophosphatidylcholine. Overproduction or suppression of G2A results in the enhancement or reduction of outgrowth induced by the treatement with $sPLA_2$ and lysophosphatidylcholine treatment. However, to our knowledge, mammalian $sPLA_2$ has not yet been reported to contribute to the neuritogenesis in the central nervous system. Rather than $sPLA_2$, $cPLA_2$-IV and $iPLA_2$-VI have been reported to be involved in the neuritogenesis in the central nervous system [23].

Bee venom $sPLA_2$-III also promotes the processes of PC 12 cells [21], but the enzyme induces neuronal apoptosis [24]. Snake venom $sPLA_2$-I or II also promotes the processes of PC 12 cells [25], but the enzyme induces neuronal apoptosis [26]. Furthermore, mammalian $sPLA_2$-IB [27] and $sPLA_2$-IIA [28]

Table 2 Neuronal apoptosis and neuritogenesis by $sPLA_2$s

PLA_2	Neuron	DRG	PC12
Snake venom $sPLA_2$-IA	Apoptosis [26]		Neuritogenesis [25]
Mammalian $sPLA_2$-IB	Apoptosis [70]		Not detected [22]
Mammalian $sPLA_2$-IIA	Apoptosis [28]		Not detected [22]
Mammalian $sPLA_2$-IIC			
Mammalian $sPLA_2$-IIE			
Mammalian $sPLA_2$-IIF			
Mammalian $sPLA_2$-III		Neuritogenesis [14]	Neuritogenesis [14]
Bee venom $sPLA_2$-III	Apoptosis [24]		Neuritogenesis [21]
Mammalian $sPLA_2$-V			Neuritogenesis [21]
Mammalian $sPLA_2$-X			Neuritogenesis [21]
Mammalian $cPLA_2$-IV	Neuritogenesis [23]		
Mammalian $iPLA_2$-VI	Neuritogenesis [23]		

Neurons include cortical neurons in the central nervous system. DRG is dorsal root ganglion in the peripheral nervous system. Apoptosis was not detected in the PC12 cells treated with mammalian $sPLA_2$-IB and $sPLA_2$-IIA

induce neuronal apoptosis, but they do not promote the processes of PC 12 cells [21, 22]. Since neurons and PC12 cells are differentiated and proliferative cells, respectively, it is no wonder that results obtained from neurons are not in accordance with those from PC12 cells. Thus, the neuritogenic effect of $sPLA_2$ should be confirmed in the central nervous system.

Neurotransmission

$sPLA_2$-IIA is contained in rat brain synaptosomes and released from the synaptosomes in response to depolarization evoked by a high concentration of potassium in the presence of Ca^{2+} [29]. When neuronally differentiated PC12 cells were stimulated with carbamylcholine or potassium, $sPLA_2$-IIA is released into the medium. $sPLA_2$-IIA inhibitors suppress catecholamine secretion from the carbamylcholine-activated PC12 cells. Exogenous $sPLA_2$-IIA alone elicits catecholamine secretion [29]. Furthermore, the localization of mature/cleaved form of $sPLA_2$-III in postsynaptic structures suggests a physiological role of the enzyme in neurotransmission or synaptic plasticity [30].

The activities of $cPLA_2$ and $sPLA_2$ increase upon exposure of cortical neurons to glutamate [31]. These studies support a functional link between PLA_2 activity and stimulation of glutamate receptors. Furthermore, $sPLA_2$ shows a synergistic effect on the increase of transient Ca^{2+} in hippocampal neurons [32], and the release of arachidonic acid (AA) in primary cortical neurons [33] induces by non-toxic and toxic glutamate concentrations. In primary cortical neurons, the combination of exogenous $sPLA_2$ and glutamate also potentiates the release of AA from phosphatidylcholine and phosphatidylethanolamine [34].

Long-Term Potentiation

Long-term potentiation (LTP) has been proposed as one of mechanisms for memory formation, and is associated with amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) receptors activation and release of AA [35]. Influx of calcium ions through *N*-methyl-D-aspartate (NMDA) receptors into the postsynaptic spine triggers biochemical processes associated with LTP, which is believed to underlie memory formation in the CNS including the hippocampus and amygdalae (Fig. 1). The increased calcium concentration in the spine activates key

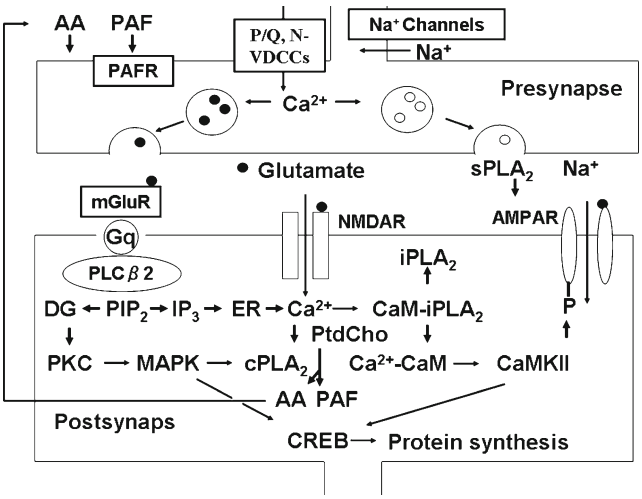

Fig. 1 Physiological roles of PLA_2 on the synaptic plasticity. Calcium plays a well-defined role in the biochemical transduction of signals from the synapse to the nucleus. In response to synaptic activity and neurotransmitter release, extracellular calcium flows into the postsynaptic cell through synaptic and extrasynaptic ligand- and voltage-gated calcium channels. Major routes of entry with well-established effects on nuclear gene expression are the NMDA receptor (*NMDAR*). Calcium-permeable AMPA receptors (*AMPAR*) may play a role at developing synapses or after the induction of synaptic plasticity. Calcium signals can also be amplified by calcium-induced release of calcium from intracellular stores, triggered by activation of inositol triphosphate receptors (IP_3R). Calcium at the mouth of the channel, in the cytoplasm, or within the nucleus can signal to activity-dependent transcription factors. Alterations in calcium influx into the postsynaptic cell during development or as a result of mutation modulate the induction of gene expression in response to neuronal activity. ER denotes endoplasmic reticulum

enzymes such as CaMK II [36] and MAPK [37]. AMPA receptors are activated by phosphorylation by CaMKII, resulting in an increase of Ca^{2+} entry into postsynaptic neurons.

PLA_2 is distributed in synaptic vesicles, and its activity is neutralized completely by an antibody raised against $sPLA_2$-IIA [29]. $sPLA_2$-IIA immunoreactive with anti-$sPLA_2$-IIA antibody is released from synaptosomes in response to depolarization evoked by a high concentration of potassium in the presence of Ca^{2+}. In rat brain membranes, bee venom $sPLA_2$-III produces a significant increase in the binding of [^{3}H]AMPA to the AMPA/quisqualate receptor. $sPLA_2$ increases the affinity of the AMPA/quisqualate receptor without changing the maximum number of sites. In contrast, $sPLA_2$ does not detectably modify the binding of [^{3}H]kainate to the kainate receptor and of [^{3}H]glutamate and [^{3}H]glycine to the NMDA receptor complex [38]. The increased sensitivity of the AMPA/quisqualate receptors contributes to the expression of LTP in area CA1 of hippocampus. NMDA receptor activation increases AA release as a result of calcium influx through the NMDA receptor channel and thereby stimulation $cPLA_2$ [39].

PLA_2 inhibitors block LTP expression in the CA1 area of hippocampal slices [40] and AA [41] or its metabolites, PAF [42, 43], have been proposed as potential messenger signals released by postsynaptic cells to modify transmitter release in LTP. Exogenously applied AA facilitates LTP formation induced by high-frequency stimulation, and AA applied during low-frequency stimulation may result in slowly developing LTP [44]. In hippocampal slices, PAF induces a stable and concentration-dependent increase in LTP [43]. The PAF effect is blocked by the PAF receptor antagonists [42, 43]. Furthermore, observations that AA- [45] and PAF-induced LTP [43] are blocked by an NMDA receptor antagonist suggest that AA and PAF do not act as a retrograde messenger, but rather they likely serves to modify the functional properties of both NMDA and AMPA subtypes of glutamate receptor [46].

A potent and selective inhibitor of $iPLA_2$, bromoenol lactone (BEL), completely suppresses the induction of LTP, suggesting a pivotal role of $iPLA_2$ in the induction of LTP [35]. Although $iPLA_2$ liberates linoleic acids from phosphatidylcholine, linoleic acid affected neither the induction nor maintenance of LTP [47]. PLA_2 serves as a key enzyme to produce free fatty acids including docosahexaenoic acid (DHA). Although the content of DHA is slightly lower than AA in the central nervous system, DHA is also one of the major components of membrane phospholipids in the brain and is a good substrate of PLA_2 partly because it is mainly localized in the 2-position of the phospholipids, particularly in phosphatidylethanolamine [48]. Therefore, as a result of the activation of PLA_2, DHA may be predominantly released and accumulated in extracellular spaces. Thus, DHA may possibly contribute to the induction of LTP. Products of PLA_2 may modulate glutamate release, postsynaptic receptor activation, and presynaptic responses [46]. In addition to lipid mediators (i.e., AA, eicosanoids and PAF), the released $sPLA_2$ from the presynaptic neuron may directly participate in the induction and/or maintenance of LTP through a receptor-mediated mechanism or alternatively through its uptake by the presynaptic neuron [49].

Pathological Roles of Secretory Phospholipase A_2 in Neurological Diseases

Alzheimer's Disease

AD is a neurodegenerative disease characterized primarily by cognitive impairment and secondarily by motor dysfunction. AD is the most common cause of late life dementia [50]. Although most cases of AD are thought to be sporadic, the risk factors for AD include stroke [51]. As well as stroke [52], $sPLA_2$ is expressed in the brain of AD patients [53]. All these conditions have a vascular involvement and reduce cerebral perfusion. AD is defined pathologically by extracellular neuritic plaques comprised of fibrillar deposits of a 4-kDa hydrophobic polypeptide known as amyloid β (Aβ) and neurofibrillary tangles (NFTs) consisting of paired helical filaments of hyperphosphorylated tau (Fig. 2). Other pathologic hallmarks of AD are activated microglia, reactive astrocytes and neuronal cell loss [51].

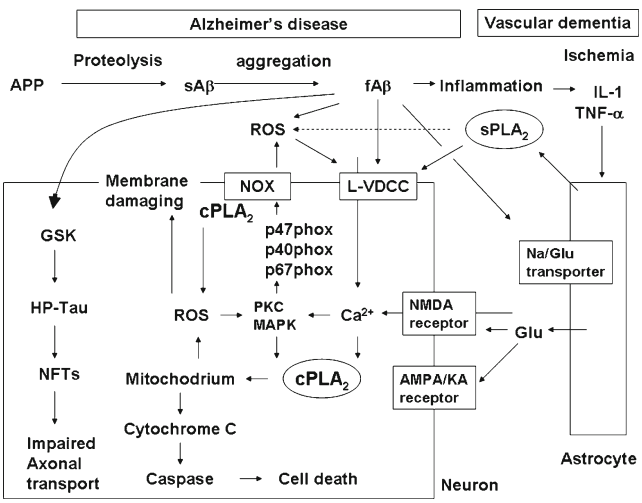

Fig. 2 Steps involved in neurodegeneration of Alzheimer's disease (*AD*) and cerebrovascular disease (*CVD*). Scheme showing the Aβ hypothesis in AD, which is based on the role of arachidonate cascade. *AMPA/KAR* *N*-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid/kainate receptor, *APP* amyloid precursor protein, *cPLA*$_2$ cytosolic phospholipase A_2, *fAβ* fibrillar amyloid β protein, *GSK* glycogen synthase kinase, *HP-Tau* hyperphosphorylated tau, *MAPK* mitogen-activated protein kinase, *Na*$^+$*/GluT* Na^+-dependent glutamate transporter, *NFTs* neurofibrillary tangles, *NMDAR* *N*-methyl-D-aspartate receptor, *Aβ* amyloid β protein oligomers, *PL* phospholipids, *ROS* reactive oxygen species, *sAβ* soluble amyloid β protein, *Glu* glutamate, *sPLA*$_2$ secretory phospholipase A_2, *sPLA*$_2$*R* secretory phospholipase A_2 receptor

The Ca^{2+} hypothesis suggests that intracellular calcium increase in response to Aβ oligomer formation may lead to neuronal cell dysfunction and death in AD [54]. As shown in Fig. 5, mechanisms that perturb normal neuronal Ca^{2+} homeostasis include aberrant Ca^{2+} influx through plasma membrane channels including L-VDCCs [55] and NMDA receptors [56]. Although soluble Aβ exhibits low toxicity, fibrillar Aβ (fAβ), an Aβ conformation similar to that found in the AD brain, greatly increase Aβ toxicity in neuronal cultures [57]. $sPLA_2$-IIA mRNA is up-regulated in AD brains as compared to non-demented elderly brains [53]. A higher percentage of $sPLA_2$-IIA-immunoreactive astrocytes present in AD hippocampus and inferior temporal gyrus. In inferior temporal gyrus, the majority of $sPLA_2$-IIA-positive astrocytes are associated with Aβ-containing plaques. Studies with human astrocytes in culture demonstrated the ability of oligomeric Aβ1-42 and interleukin-1β (IL-1β) to induce $sPLA_2$-IIA mRNA expression, indicating that this gene is among those induced by inflammatory cytokines (Fig. 2).

fAβ [55] and $sPLA_2$-IIA [58] generate reactive oxygen species (ROS), which cause membrane lipid peroxidation and disturb the integrity of neuronal membranes (Fig. 2). These free radicals stimulate L-VDCC [59], and potentiate the influx of Ca^{2+} into neurons [55, 58]. Increased oxidative stress is an early event in AD that decreases with disease progression and formation of characteristic lesions [60]. Products of lipid peroxidation show a significant increase of intracellular Aβ production [61]. Aβ causes neuronal apoptosis via the activation of caspase [62]. $sPLA_2$-IB exerts its apoptotic activity via the release of cytochrome *c*, which results in triggering the activation of caspase cascade and poly(ADP-ribose) polymerase cleavage [63].

Cerebrovascular Disease

CVD is caused by vascular lesions, cerebral infarctions, multiple lacunar infarctions and ischemic periventricular leukoencephalopathy. CVD can result from ischemic or hemorrhagic brain damage. The three most common mechanisms causing this disease are single, strategically placed infarcts; multiple cortical infarcts; and subcortical small-vessel disease [64]. Stroke, the potent risk factor for CVD, is caused by a critical alteration of blood flow to a region of the brain [65]. An acute obstruction of an artery results in ischemia, i.e., insufficient blood flow to the tissue [66]. The ischemic brain suffers from a mismatch between its cellular energy demands and the ability of the vascular system to supply substrate, most importantly, oxygen. Subsequently, neurologic malfunctions and neuronal cell death are caused by increased intracellular calcium, excessive extracellular glutamate, free radicals, and inflammation (Fig. 2). At the beginning of the stroke, there is a definite gradation of injury — a central area or core, with low blood flow already showing signs of massive cell death, and an outer area, the penumbra, that is still alive, but will malfunction after several days. A rat with the middle cerebral artery occlusion has been established as an animal model for stroke [67]. The occlusion causes irreversible necrosis and infarction in the core [68]. On the other hand, cell death is induced not only via necrosis, but also via apoptosis, and cells remain viable for several hours in the penumbra [69].

PLA_2 inhibitors terminate the reversible proapoptotic state in the penumbra. Therefore, interventions designed to terminate the reversible proapoptotic state are expected to reduce the ischemic damage and lead to successful treatment of stroke. The cortical $sPLA_2$ is induced in response to focal cerebral ischemia [27, 28, 52, 70]. $sPLA_2$ activity is elevated in the cortex, in which the ischemic core and the penumbra coexist [28]. $sPLA_2$-IB [71] and $sPLA_2$-IIA [58] induces neuronal apoptosis via L-VDCC (Fig. 2). A few minutes after uncompensated brain ischemia, cell death pathways overcome survival-promoting pathways, leading to neuronal death through three interacting mechanisms: excitotoxicity attributable to excess glutamate, oxidative stress, and/or stimulation of apoptotic-like pathways [51].

Multiple Sclerosis and Experimental Autoimmune Encephalomyelitis

The pathology of multiple sclerosis involves both antigen specific mechanisms and the innate immune system, including elements of the acute inflammatory response [72]. Since axonal loss is likely to begin at disease onset, the inflammation that accompanies this degeneration may be a persistent contributing factor in multiple sclerosis, as it all disorders in which there is destruction of nervous tissue. Rodent experimental autoimmune encephalomyelitis models of multiple sclerosis suggest PLA_2 enzymes are involved in the onset and genesis of this disease [73]. Systemic infusion of anchored lipid conjugates, targeting $sPLA_2$s, attenuates aspects of the autoimmune response and experimental autoimmune encephalomyelitis clinical disease. Multiple Sclerosis patients also show elevations in $sPLA_2$ enzyme activity [74]. Enzymatically active $sPLA_2$ in the urine increases following immunization and peaked between days 8 and 10 PI, which is just prior to the onset of experimental autoimmune encephalomyelitis symptoms [74].

Spinal Cord Injury

Spinal cord injury starts with the primary injury phase, characterized by an initial damage core generally induced by the application of mechanical forces [75]. The secondary injury phase is characterized by the activation of a complex network of cellular processes, such as microglial and astroglial reactivity, leukocyte infiltration and mobilization of neural precursors. The activation of the secondary injury phase usually leads to the massive death of neural cells via apoptosis as well

as necrosis and to the disruption of neural circuits that surround the primary injury core, and these cause most of the functional deficits associated with spinal cord injury [75]. The neurochemical changes include a rise in glutamate and intracellular calcium, degradation of membrane phospholipids with generation of free fatty acids, diacylglycerols, eicosanoids, and lipid peroxides, and activation of phospholipases and lipases [2]. After the initial mechanical insult of spinal cord injury, secondary mediators propagate a massive loss of oligodendrocytes. Following spinal cord injury, both the total phospholipase activity and cytosolic $cPLA_2$ α protein expression are increased. Cervical spinal cord injury markedly upregulated mRNA of $sPLA_2$-IIA and IIE. In contrast, spinal cord injury induces down regulation of $sPLA_2$-X, and no change in $sPLA_2$-IB, IIC, V, and XIIA expression. At the lesion site, $sPLA_2$-IIA and IIE expression are localized to oligodendrocytes. $sPLA_2$-IIA exhibit cytotoxicity in differentiated adult oligodendrocyte precursor cells but not primary astrocytes or Schwann cells [76].

Epilepsy

In the pentylenetetrazol-induced model of epilepsy, $sPLA_2$ activity is significantly elevated in cortical, hippocampal, and cerebellar regions compared with the control group in rat brain. The increase in $sPLA_2$ activity is more pronounced in hippocampal and cortical regions than in the cerebellar region [77]. PLA_2 activity is increased in the brain, as frequently reported in schizophrenia, is also related to psychosis in epilepsy. The prevalence of schizophrenia-like psychosis in patients with temporal lobe epilepsy secondary to mesial temporal sclerosis is higher than that in patients with other forms of epilepsy. Temporal lobe epilepsy secondary to mesial temporal sclerosis in patients with psychosis shows a significantly higher brain $iPLA_2$ activity as compared to patients without psychosis. No significant differences were found between both groups regarding $sPLA_2$ and $cPLA_2$ activities. Epileptic seizures are known to stimulate $cPLA_2$ activity and its expression with the accumulation of AA [78]. More studies are needed on the involvement of PLA_2 isoforms in the pathogenesis of epilepsy.

Pathological Functions of Phospholipase A_2

Secretory Phospholipase A_2 Receptor

Certain types of snake venom $sPLA_2$ have specific binding proteins similar to the mannose receptor in mammals [79]. Two main types of high affinity receptors for Taipan snake venom $sPLA_2$s are identified as N-type and M-type (Fig. 3). N-type $sPLA_2$ receptors are first identified in rat brain membranes [80]. These receptors have high affinities for neurotoxic

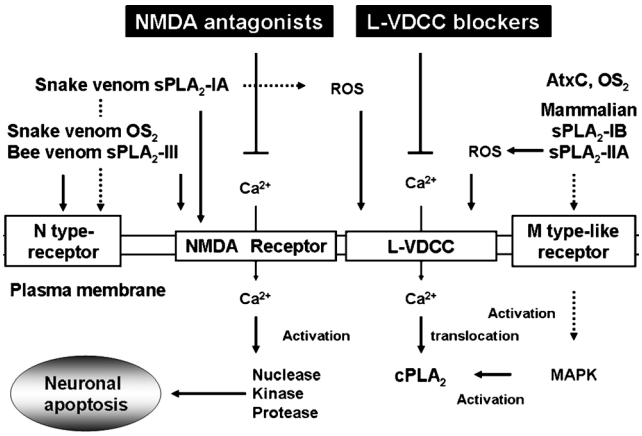

Fig. 3 Secretory phospholipase A_2 receptors. Mammalian $sPLA_2$s generate ROS, open L-VDCCs, but not glutamate receptors and incorporates Ca^{2+} into neurons. Snake venom $sPLA_2$s also generates ROS, opens L-VDCCs, activate NMDA receptors and uptake Ca^{2+} into neurons. The elevated level of intracellular calcium concentration activates Ca^{2+}-dependent enzymes such as $cPLA_2$, nucleases, kinases and proteases, and induced neuronal apoptosis

$sPLA_2$s such as OS_2 and the bee venom $sPLA_2$ ($sPLA_2$-III) but not for nontoxic $sPLA_2$s such as OS_1, suggesting that N-type receptors contribute to the neurotoxic effects of venom $sPLA_2$s. M-type $sPLA_2$ receptors are first identified in rabbit skeletal muscle cells. In porcine cerebral cortex, a receptor for long-nosed viper venom ammodytoxin C (AtxC) is identified [81]. In its molecular mass and pharmacological profile, the AtxC receptor resembles the M-type receptor for $sPLA_2$ from rabbit skeletal muscle (a C-type multilectin, homologous to macrophage mannose receptor), yet in terms of relative abundance in brain and antigenicity, these two receptors are completely different. The AtxC receptor consists of a single 180-kDa subunit and recognizes with high affinity OS_2 but not the neurotoxic $sPLA_2$-III. On the other hand, ^{125}I-labeled $ppPLA_2$-IB has revealed two classes of high-affinity binding sites in the rat brain [82]. Their binding profile is different from that of binding sites of OS_2, suggesting the existence of a novel class of $sPLA_2$ N-type binding sites.

Mammalian $sPLA_2$ can only bind to the M-type receptor, which is not restricted to muscles but also expressed in various tissues [83]. Mammalian $sPLA_2$-IB are not only digestive enzymes, but also involved in a number of other important physiological processes such as cell contraction [84], lipid mediator release [85] and cell proliferation [86] via its specific receptors. The receptor of porcine pancreatic $sPLA_2$-IB ($sPLA_2R$) is composed of a single glycosylated polypeptide chain with an apparent molecular mass of 190 kDa [87]. The $sPLA_2R$ for porcine pancreatic $sPLA_2$-IB is structurally related to the macrophage mannose receptor, a unique member of Ca^{2+}-dependent (C-type) animal lectin family [88]. The $sPLA_2$-IB receptor carries a region consisting of eight tandem carbohydrate-recognition-domain-(CRD)-like domains as in the case of the mannose receptor; this region is responsible

for $sPLA_2$-IB binding, not sugar binding, in the $sPLA_2$-IB receptor. Although the $sPLA_2R$ has not yet been detected in the CNS, $sPLA_2$-IB is distributed in rat brain and human brain [7].

Secretory PLA_2-Induced Neuronal Cell Death

Porcine pancreatic $sPLA_2$-IB induces neuronal cell death in primary cultures of rat cortical neurons [70]. $sPLA_2$-IB shrinks neuronal cell bodies and shortens neurites. In the early stage of cell death, it becomes difficult to resolve the plasma membrane, whereas features in the cytosol and the nucleus are unaltered. In the middle stage, microtubules, neurofilaments, and ribosomes are condensed as the neurons continued to shrink. Moreover, a decrease of the rough endoplasmic reticulum (ER) and progressive swelling of the Golgi cisternae are observed within the cytoplasm. The nuclei shrink progressively, and chromatin clumps become increasingly electron-dense. In the late stage, intracellular organelles such as the ER and the Golgi apparatus are lost, but the mitochondria remained intact. Condensation and fragmentation of chromatin are noted in the nucleus. The dying cells finally are fragmented into small pieces. The $sPLA_2$-IB-induced neuronal cell death exhibits apoptotic features with blebbed membrane, condensed chromatin, and fragmented DNA. The ultrastructural disruption in $sPLA_2$-IB-treated neurons is suppressed completely by a $sPLA_2$ inhibitor, indoxam [70].

Porcine pancreatic $sPLA_2$-IB induces neuronal apoptosis via its high-affinity binding sites in the rat brain [27]. The specific binding sites of [^{125}I]$sPLA_2$-IB are recognized by an anti-$sPLA_2R$ antibody. The anti-$sPLA_2R$ IgG also causes neuronal cell death as well as $sPLA_2$-IB. By Scatchard analysis, number of maximal binding sites (B_{max}) and dissociation constant (K_d) values of [^{125}I]$sPLA_2$-IB is 3.1 fmol/10^6 cells and 0.84 nM, respectively [27]. Although 40 % of [^{125}I]$sPLA_2$-IB bound to neurons is displaced by 100 nM $sPLA_2$-IB, bee venom $sPLA_2$-III, a potent inhibitor of N-type binding sites of OS_2 [80], significantly increases the binding of [^{125}I]$sPLA_2$-IB to neurons. The specific binding sites of [^{125}I]$sPLA_2$-IB are distinguishable from the N-type binding sites. $sPLA_2$-IB appears to cause neuronal cell death via the specific binding sites, which are different from the $sPLA_2R$ and the N-type binding sites of OS_2.

Human recombinant and rat spleen $sPLA_2$-IIA causes disruption of neurites and cell bodies rather than their shrink in a different fashion to $sPLA_2$-IB. However, as evidenced by condensed chromatin and fragmented DNA [28], ultrastructural characteristics of neuronal cell death by $sPLA_2$-IIA are apoptotic. The M-type receptor binds toxic and non-toxic $sPLA_2$ of either group I or II [81], giving rise to a possibility that $sPLA_2$-IB and $sPLA_2$-IIA might induce neuronal apoptosis via the AtxC receptor. The above two $sPLA_2$s-induced neuronal apoptosis is suppressed by a reversible $sPLA_2$ inhibitor, indoxam, as well as a putative irreversible $sPLA_2$ inhibitor [28, 70]. Under the optimal conditions of each $sPLA_2$ reaction with 1-palmitoyl-2-oleoyl-*sn*-glycero-3- phosphocholine as a substrate, IC_{50} values of indoxam against $sPLA_2$-IIA, -IID, and -IIE are 1–2 nM [89]. On the other hand, the other isoforms ($sPLA_2$-IB, -V, and -X) are less sensitive to indoxam with over 100-fold higher IC_{50} values. Indoxam does not only inhibit the enzymatic activity of $sPLA_2$s, but also blocks the binding to its murine receptor (K_i=30 nM) [89], suggesting that $sPLA_2$s might cause apoptosis via its high affinity binding sites and its enzymatic activity.

$sPLA_2$-Induced Neuronal Apoptosis via L-type Voltage-Dependent Ca^{2+} Channels

Calcium is an intracellular second messenger in many cell types, and regulates many complicated cellular processes, including cell activation, proliferation and apoptosis. In resting cells, the intracellular Ca^{2+} concentration ([Ca^{2+}]i) is maintained at a low level (50–150 nM) compared with the levels present in the extracellular space (2 mM). However, agonists including hormones, growth factors and neurotransmitters induce changes in the intracellular Ca^{2+} dynamics, including elevation of [Ca^{2+}]i via mobilization of Ca^{2+} from intracellular stores such as the ER and mitochondria as well as the extracellular fluid. The calcium chelator attenuates the neurotoxicity of mammalian and non-mammalian $sPLA_2$s, suggesting that calcium mobilization might contribute to the apoptosis. Actually, these $sPLA_2$s elevate the influx of Ca^{2+} persistently before apoptosis [58, 71].

Neuronal cell death is induced synergistically by non mammalian $sPLA_2$s (bee venom $sPLA_2$ and Taipan snake venom $sPLA_2$-OS_2) glutamate in rat cortical neurons [33]. Glutamate receptors have been classified as ionotropic and metabotropic. Ionotropic glutamate (AMPA, kainate and NMDA) receptors are ligand-gated cation channels, whereas metabotropic glutamate receptors are coupled to various signal transduction systems. NMDARs are unique in that their activation is governed by a strong voltage-dependence due to receptor channel blockade by Mg^{2+} at hyperpolarized membrane potentials. Mg^{2+} blockade gives NMDA receptors their characteristic negative slope conductance.

Snake venom $sPLA_2$s are neurotoxic in vivo and in vitro, causing synergistic neurotoxicity to cortical cultures when applied with toxic concentrations of glutamate [33]. Antagonists for NMDA receptor and AMPA receptor prevent neurons from the Tipan snake venom $sPLA_2$-OS_2-induced neuronal cell death [90]. Snake venom $sPLA_2$-IA also induces neuronal cell death via apoptosis. $sPLA_2$-IA disrupts cell bodies and fragmented neurites, accompanied with chromatin condensation and DNA fragmentation [26]. NMDA receptor blockers such as PCP and DCKA, but not AMPA receptor blocker, prevent neurons from undergoing the $sPLA_2$-IA-induced neuronal cell death (Fig. 4).

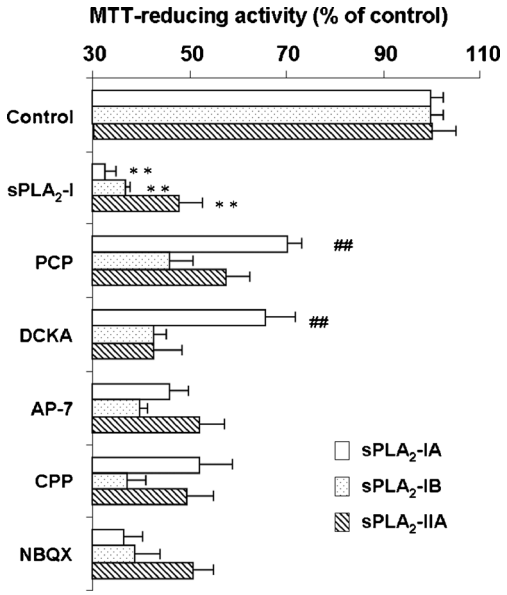

Fig. 4 NMDA receptor blockers rescued neuron from undergoing the $sPLA_2$s-induced neuronal cell death. Glutamate receptor blockers: cortical neurons were treated with 20 pM $sPLA_2$-IA (*open columns*), 50 nM $sPLA_2$-IB (*dotted columns*) or 1 μM $sPLA_2$-IIA (*hatched columns*) in the absence or presence of 30 μM PCP, DCKA, AP-7, CPP or NBQX. MTT reducing activity was determined 24 h ($sPLA_2$-IA and $sPLA_2$-IB) or 48 h ($sPLA_2$-IIA) later. Data are expressed as means±SEM ($n=4$). $^{**}P<0.01$, compared with control; $^{\#}P<0.05$, $^{\#\#}P<0.01$, compared with $sPLA_2$-IA or $sPLA_2$-IB alone by ANOVA followed by Dunnett's test

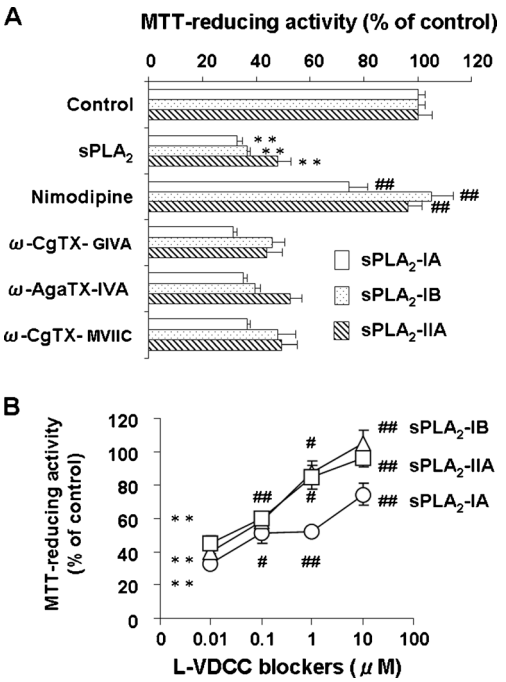

Fig. 5 L-VDCC blockers rescued neuron from undergoing the $sPLA_2$s-induced neuronal cell death. **a** VDCC blockers: Cortical neurons were treated with 20 pM $sPLA_2$-IA (*open columns*), 50 nM $sPLA_2$-IB (*dotted columns*) or 1 μM $sPLA_2$-IIA (*hatched columns*) in the absence or presence of 10 μM Nimodipine, 1 μM ω-Aga GVIA, ω-CgTX GVIA or ω-CgTX-MVIIC. **b** Concentration: cortical neurons were treated with S-312d or nimodipine at the indicated concentrations in the presence of 20 pM $sPLA_2$-IA (*circles*), 50 nM $sPLA_2$-IB (*triangles*) or 1 μM $sPLA_2$-IIA (*squares*). MTT reducing activity was determined 24 h ($sPLA_2$-IA and $sPLA_2$-IB) or 48 h ($sPLA_2$-IIA) later. Data are expressed as means± SEM ($n=4$). $^{**}P<0.01$, compared with control; $^{\#}P<0.05$, $^{\#\#}P<0.01$, compared with $sPLA_2$-IA or $sPLA_2$-IB or $sPLA_2$-IIA alone by ANOVA followed by Dunnett's test

However, neither NMDA receptor nor AMPA/kinate receptor are involved in the neurotoxicity of mammalian $sPLA_2$-IB (porcine pancreas) and $sPLA_2$-IIA (human recombinant) [58, 71] (Fig. 4). Moreover, neither N-type nor P/Q-type VDCCs affects the mammalian and non-mammalian $sPLA_2$s-induced neuronal cell death (Fig. 5A). An L-type voltage-dependent Ca^{2+} channels (L-VDCC) blocker, nimodipine, completely prevent neurons from undergoing the mammalian $sPLA_2$IB- and $sPLA_2$IIA-induced cell deaths, whereas it does partially neurons from undergoing the non-mammalian cobra venom $sPLA_2$IA (*Naja naja*)-induced cell death (Fig. 5B). The elevation of Ca^{2+} uptake by these $sPLA_2$s is significantly reduced by L-VDCC blockers [26, 58, 71]. Neither an N-type VDCC blocker nor P/Q-type VDCC blockers affects $sPLA_2$-increased Ca^{2+} influx. L-VDCC blockers ameliorate mammalian and non-mammalian $sPLA_2$s-induced morphological changes as well as biochemical apoptotic features [26, 58, 71]. Thus, mammalian $sPLA_2$s induce neuronal apoptosis via the influx of Ca^{2+} into neurons through L-VDCC, whereas non-mammalian $sPLA_2$ does through the glutamate receptor and the L-VDCC (Fig. 3).

The L-VDCC is activated by ROS [55], which is generated by $sPLA_2$-IIA before neuronal cell death [58]. ROS are reduced significantly by radical scavengers, including vitamin E, probucol, butylated hydroxytoluene and chlorpromazine. On the other hand, nimodipine does not affect the production of ROS after $sPLA_2$-IIA treatment. Vitamin E also suppresses Ca^{2+} influx as well as the L-VSCC blocker. In addition, the above radical scavengers significantly prevent neurons from undergoing $sPLA_2$-IIA-induced neuronal cell death [58]. As the common pathway, the excess influx of calcium via L-VDCC contributes the neurotoxicity of mammalian and non-mammalian $sPLA_2$s (Fig. 3).

Pathological Calcium Signaling

VDCCs play pathophysiological roles in coordination with the LGCCs and the intracellular stores (Fig. 2). The elevation of $[Ca^{2+}]i$ activation Ca^{2+}-dependent enzymes such as protein kinases, proteinases, DNases, and PLA_2s. Calcium mediated activation of calpain is also reported to cause cell death in various neuropathological conditions including CVD and AD [91]. The requirement for calcium and protein kinases (MAPK and PKC) for $cPLA_2$ suggests that $cPLA_2$ is under tight

control by intracellular signaling pathways. The elevation of $[Ca^{2+}]i$ through L-VDCC and NMDA receptor activates PKC, MAPK ad $cPLA_2$ (Fig. 2). Moreover, it is linked with the production of ROS and apoptosis through activation of the superoxide producing enzyme NADPH oxidase (NOX). There are at least seven known isoforms, NOX1-5, Duox1 and 2, each with a unique combination of subunits [92]. NOX2 is well studied in phagocytic cells, macrophages, and endothelial cells and is comprised of the subunits p47phox, p67phox, p40phox, and Rac 1 in the cytosol and gp91phox and p22phox in the membrane fraction (plasma membranes or other subcellular membranes). Activation of NOX2 is dependent on phosphorylation of the cytosolic subunits, e.g., phosphorylation of p47phox by protein kinase C in human monocytes [93]. Subsequently, the cytosolic subunits form a complex and translocated to the membrane-associated gp91phox subunit. NOX1, 2, and 4 are expressed in neurons, astrocytes, and microglia in the CNS [92]. NOX is involved in excitotoxicity induced by ionotropic glutamate receptors, including the NMDA subtype [94]. NMDA-induced activation of NOX can trigger signaling pathways leading to activation of ERK1/2, the protein kinase required for activation of $cPLA_2$ (Fig. 2). Oligomeric Aβ can induce ROS production through NOX. In turn, ROS from NOX can stimulate ERK1/2 phosphorylation and activation of $cPLA_2$ and result in a release of AA [95]. The excitotoxic effect of Aβ is inhibited by NMDA receptor antagonists, including memantine, a drug used to treat AD patients [96].

Conclusions

$sPLA_2$s cause neuritogenesis in peripheral neurons, whereas $cPLA_2$s and $iPLA_2$s — rather than $sPLA_2$ — are involved in the neuritogenesis in central neurons. $sPLA_2$s are localized at synapse and secret catecholamine from neuronally differentiated PC12 cells, suggesting that the enzymes may release neurotransmitters from neurons. $iPLA_2$s play a pivotal role in the induction of LTP. Although linoleic acid is preferentially produced by $iPLA_2$, it does not induce and maintain LTP. Since PAF induces LTP and PLA_2 inhibitors block the induction of LTP, $sPLA_2$ released from the presynaptic neuron appears to directly participate in the induction and maintenance of LTP.

The abnormal genetic and environmental processes interfere with normal brain maturation and result in dysfunctional monoaminergic neurotransmission associated with the phenotype of schizophrenia. Genetic variation in $sPLA_2$-IIA, $cPLA_2$-IVA and $iPLA_2$-VIA is possibly involved in both brain maturational processes and neurotransmission [97]. The elevation of PLA_2 activity is correlated with structural changes in schizophrenic brains [98]. The increased activity of $iPLA_2$ in the brain of schizophrenic patients has been suggested to accelerate the breakdown of membrane phospholipids and alter the properties of neuronal membranes, which in turn contribute to a hypodopaminergy [99]. A genetic variant of $cPLA_2$ gene has been reported to increase risk for schizophrenia through an increment of PLA_2 activity [100]. Since the elevation of PLA_2 activity is correlated with structural changes in schizophrenic brains [98], its activity and protein may be altered in the disease.

The importance of PLA_2 in the pathogenesis of the neuronal degeneration in prion diseases has been indicated by the use of PLA_2 inhibitors that reduced the misfolding of the normal prion protein, caspase-3 activity and prostaglandin E_2 production [101]. Since $cPLA_2$ plays a vital role in the progress of prion diseases [102], $sPLA_2$ might be also associated to the prion diseases. $iPLA_2$ is the causative gene for early-onset PARK14-linked dystonia–parkinsonism [103]. Mice deficient in $cPLA_2$ activity are resistant to 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine neurotoxicity, suggesting that $cPLA_2$ is closely associated with the pathophysiology of Parkinson's diseases [104]. A PLA_2 inhibitor, quinacrine, protects do dopaminergic neurons from neurodegeneration in the 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine-induced model of parkinsonism [105], suggesting the plausible involvement of $sPLA_2$ in Parkinson's diseases.

$sPLA_2$s are up-regulated in the cerebrovascular disease, stroke, and the neurodegenerative diseases, AD, in which the neuronal apoptosis is the common pathology. $sPLA_2$s induce neuronal apoptosis via L-VDCC, but not via NMDAR, whereas non-mammalian $sPLA_2$s induce neuronal apoptosis via L-VDCC and NMDAR [26]. Although mammalian $sPLA_2$s bind M type-like receptor in the brain, it has not yet been clarified how M type-like receptor contributes to the apoptosis. To this end, the neurotoxicity of $sPLA_2$s is needed to be analyzed in the M type-like receptor knock-out mouse. Among arachidonate metabolites, 15-deoxy-$\Delta^{12,14}$-PGJ_2 can be contributed to the neurotoxicity of $sPLA_2$s. Prior to neuronal apoptosis, $sPLA_2$s enhance the influx of Ca^{2+} and produce PGD_2. Although PGD_2 receptors have not yet been detected on the neuronal surface, PGD_2 can be non-enzymatically metabolized to 15-deoxy-$\Delta^{12,14}$-PGJ_2. 15-deoxy-$\Delta^{12,14}$-PGJ_2 possesses dual functions as a neuroprotectant and a neurotoxicant. 15-deoxy-$\Delta^{12,14}$-PGJ_2 has been reported to exhibit the neuroprotective effect via its nuclear receptor, peroxysome proliferators-activated receptor γ [106] and the neurotoxic effect via its specific binding sites in the plasma membrane of neurons [107]. One of problems to be resolved is whether the receptor binding and/or the enzymatic activity are required to the neurotoxicity of $sPLA_2$s.

Conflicts of interest The authors have declared that no competing interests exist.

References

1. Goracci G, Ferrini M, Nardicchi V (2010) Low molecular weight phospholipases A2 in mammalian brain and neural cells: roles in functions and dysfunctions. Mol Neurobiol 41(2–3):274–289
2. Farooqui AA, Ong WY, Horrocks LA (2006) Inhibitors of brain phospholipase A2 activity: their neuropharmacological effects and therapeutic importance for the treatment of neurologic disorders. Pharmacol Rev 58(3):591–620
3. Laychock SG (1982) Phospholipase A2 activity in pancreatic islets is calcium-dependent and stimulated by glucose. Cell Calcium 3(1): 43–54
4. Clapham DE (1995) Calcium signaling. Cell 80(2):259–268
5. Rosa AO, Rapoport SI (2009) Intracellular- and extracellular-derived Ca(2+) influence phospholipase A(2)-mediated fatty acid release from brain phospholipids. Biochim Biophys Acta 1791(8): 697–705
6. Grataroli R, De Caro A, Guy O, Amic J, Figarella C (1981) Isolation and properties of prophospholipase A2 from human pancreatic juice. Biochimie 63(8–9):677–684
7. Kolko M, Christoffersen NR, Varoqui H, Bazan NG (2005) Expression and induction of secretory phospholipase A2 group IB in brain. Cell Mol Neurobiol 25(7):1107–1122
8. Suzuki N, Ishizaki J, Yokota Y, Higashino K, Ono T, Ikeda M, Fujii N, Kawamoto K, Hanasaki K (2000) Structures, enzymatic properties, and expression of novel human and mouse secretory phospholipase A(2)s. J Biol Chem 275(8):5785–5793
9. Molloy GY, Rattray M, Williams RJ (1998) Genes encoding multiple forms of phospholipase A2 are expressed in rat brain. Neurosci Lett 258(3):139–142
10. Valentin E, Koduri RS, Scimeca JC, Carle G, Gelb MH, Lazdunski M, Lambeau G (1999) Cloning and recombinant expression of a novel mouse-secreted phospholipase A2. J Biol Chem 274(27): 19152–19160
11. Kolko M, Christoffersen NR, Barreiro SG, Miller ML, Pizza AJ, Bazan NG (2006) Characterization and location of secretory phospholipase A2 groups IIE, V, and X in the rat brain. J Neurosci Res 83(5):874–882
12. Valentin E, Singer AG, Ghomashchi F, Lazdunski M, Gelb MH, Lambeau G (2000) Cloning and recombinant expression of human group IIF-secreted phospholipase A(2). Biochem Biophys Res Commun 279(1):223–228
13. Valentin E, Ghomashchi F, Gelb MH, Lazdunski M, Lambeau G (2000) Novel human secreted phospholipase A(2) with homology to the group III bee venom enzyme. J Biol Chem 275(11):7492–7496
14. Masuda S, Yamamoto K, Hirabayashi T, Ishikawa Y, Ishii T, Kudo I, Murakami M (2008) Human group III secreted phospholipase A2 promotes neuronal outgrowth and survival. Biochem J 409(2):429–438
15. Chen J, Engle SJ, Seilhamer JJ, Tischfield JA (1994) Cloning and recombinant expression of a novel human low molecular weight Ca(2+)-dependent phospholipase A2. J Biol Chem 269(4):2365–2368
16. Shirai Y, Ito M (2004) Specific differential expression of phospholipase A2 subtypes in rat cerebellum. J Neurocytol 33(3):297–307
17. Nardicchi V, Macchioni L, Ferrini M, Goracci G (2007) The presence of a secretory phospholipase A2 in the nuclei of neuronal and glial cells of rat brain cortex. Biochim Biophys Acta 1771(11): 1345–1352
18. Cupillard L, Koumanov K, Mattei MG, Lazdunski M, Lambeau G (1997) Cloning, chromosomal mapping, and expression of a novel human secretory phospholipase A2. J Biol Chem 272(25):15745–15752
19. Gelb MH, Valentin E, Ghomashchi F, Lazdunski M, Lambeau G (2000) Cloning and recombinant expression of a structurally novel human secreted phospholipase A2. J Biol Chem 275(51):39823–39826
20. Negre-Aminou P, Nemenoff RA, Wood MR, de la Houssaye BA, Pfenninger KH (1996) Characterization of phospholipase A2 activity enriched in the nerve growth cone. J Neurochem 67(6):2599–2608
21. Nakashima S, Kitamoto K, Arioka M (2004) The catalytic activity, but not receptor binding, of sPLA2s plays a critical role for neurite outgrowth induction in PC12 cells. Brain Res 1015(1–2): 207–211
22. Ikeno Y, Konno N, Cheon SH, Bolchi A, Ottonello S, Kitamoto K, Arioka M (2005) Secretory phospholipases A2 induce neurite outgrowth in PC12 cells through lysophosphatidylcholine generation and activation of G2A receptor. J Biol Chem 280(30):28044–28052
23. Forlenza OV, Mendes CT, Marie SK, Gattaz WF (2007) Inhibition of phospholipase A2 reduces neurite outgrowth and neuronal viability. Prostaglandins Leukot Essent Fatty Acids 76(1):47–55
24. DeCoster MA (2003) Group III secreted phospholipase A2 causes apoptosis in rat primary cortical neuronal cultures. Brain Res 988(1–2):20–28
25. Makarova YV, Osipov AV, Tsetlin VI, Utkin YN (2006) Influence of phospholipases A2 from snake venoms on survival and neurite outgrowth in pheochromocytoma cell line PC12. Biochemistry (Mosc) 71(6):678–684
26. Yagami T, Yamamoto Y, Koma H, Nakamura T, Takasu N, Okamura N (2013) L-type voltage-dependent calcium channel is involved in the snake venom group IA secretory phospholipase A2-induced neuronal apoptosis. Neurotoxicology 35:146–153
27. Yagami T, Ueda K, Asakura K, Hata S, Kuroda T, Sakaeda T, Kishino J, Sakaguchi G, Itoh N, Hori Y (2002) Group IB secretory phospholipase A(2)induces cell death in the cultured cortical neurons: a possible involvement of its binding sites. Brain Res 949(1–2):197–201
28. Yagami T, Ueda K, Asakura K, Hata S, Kuroda T, Sakaeda T, Takasu N, Tanaka K, Gemba T, Hori Y (2002) Human group IIA secretory phospholipase A2 induces neuronal cell death via apoptosis. Mol Pharmacol 61(1):114–126
29. Matsuzawa A, Murakami M, Atsumi G, Imai K, Prados P, Inoue K, Kudo I (1996) Release of secretory phospholipase A2 from rat neuronal cells and its possible function in the regulation of catecholamine secretion. Biochem J 318(Pt 2):701–709
30. Yang H, Siddiqi NJ, Alhomida AS, Ong WY (2013) Expression and localization of sPLA2-III in the rat CNS. Neurochem Res 38(4): 753–760
31. Kim DK, Rordorf G, Nemenoff RA, Koroshetz WJ, Bonventre JV (1995) Glutamate stably enhances the activity of two cytosolic forms of phospholipase A2 in brain cortical cultures. Biochem J 310(Pt 1):83–90
32. DeCoster MA, Lambeau G, Lazdunski M, Bazan NG (2002) Secreted phospholipase A2 potentiates glutamate-induced calcium increase and cell death in primary neuronal cultures. J Neurosci Res 67(5):634–645
33. Kolko M, DeCoster MA, de Turco EB, Bazan NG (1996) Synergy by secretory phospholipase A2 and glutamate on inducing cell death and sustained arachidonic acid metabolic changes in primary cortical neuronal cultures. J Biol Chem 271(51):32722–32728
34. Rodriguez De Turco EB, Jackson FR, DeCoster MA, Kolko M, Bazan NG (2002) Glutamate signalling and secretory phospholipase A2 modulate the release of arachidonic acid from neuronal membranes. J Neurosci Res 68(5):558–567
35. Wolf MJ, Izumi Y, Zorumski CF, Gross RW (1995) Long-term potentiation requires activation of calcium-independent phospholipase A2. FEBS Lett 377(3):358–362
36. Malinow R, Schulman H, Tsien RW (1989) Inhibition of postsynaptic PKC or CaMKII blocks induction but not expression of LTP. Science 245(4920):862–866

37. English JD, Sweatt JD (1997) A requirement for the mitogen-activated protein kinase cascade in hippocampal long term potentiation. J Biol Chem 272(31):19103–19106
38. Massicotte G, Baudry M (1990) Modulation of DL-alpha-Amino-3-hydroxy-5-methylisoxazole-4-propionate (AMPA)/quisqualate receptors by phospholipase A2 treatment. Neurosci Lett 118(2):245–248
39. Dumuis A, Sebben M, Haynes L, Pin JP, Bockaert J (1988) NMDA receptors activate the arachidonic acid cascade system in striatal neurons. Nature 336(6194):68–70
40. Okada D, Yamagishi S, Sugiyama H (1989) Differential effects of phospholipase inhibitors in long-term potentiation in the rat hippocampal mossy fiber synapses and Schaffer/commissural synapses. Neurosci Lett 100(1–3):141–146
41. Drapeau C, Pellerin L, Wolfe LS, Avoli M (1990) Long-term changes of synaptic transmission induced by arachidonic acid in the CA1 subfield of the rat hippocampus. Neurosci Lett 115(2–3): 286–292
42. Arai A, Lynch G (1992) Antagonists of the platelet-activating factor receptor block long-term potentiation in hippocampal slices. Eur J Neurosci 4(5):411–419
43. Wieraszko A, Li G, Kornecki E, Hogan MV, Ehrlich YH (1993) Long-term potentiation in the hippocampus induced by platelet-activating factor. Neuron 10(3):553–557
44. Williams JH, Errington ML, Lynch MA, Bliss TV (1989) Arachidonic acid induces a long-term activity-dependent enhancement of synaptic transmission in the hippocampus. Nature 341(6244):739–742
45. O'Dell TJ, Hawkins RD, Kandel ER, Arancio O (1991) Tests of the roles of two diffusible substances in long-term potentiation: evidence for nitric oxide as a possible early retrograde messenger. Proc Natl Acad Sci U S A 88(24):11285–11289
46. Miller B, Sarantis M, Traynelis SF, Attwell D (1992) Potentiation of NMDA receptor currents by arachidonic acid. Nature 355(6362): 722–725
47. Fujita S, Ikegaya Y, Nishikawa M, Nishiyama N, Matsuki N (2001) Docosahexaenoic acid improves long-term potentiation attenuated by phospholipase A(2) inhibitor in rat hippocampal slices. Br J Pharmacol 132(7):1417–1422
48. Hadjiagapiou C, Spector AA (1987) Docosahexaenoic acid metabolism and effect on prostacyclin production in endothelial cells. Arch Biochem Biophys 253(1):1–12
49. Praznikar ZJ, Kovacic L, Rowan EG, Romih R, Rusmini P, Poletti A, Krizaj I, Pungercar J (2008) A presynaptically toxic secreted phospholipase A2 is internalized into motoneuron-like cells where it is rapidly translocated into the cytosol. Biochim Biophys Acta 1783(6):1129–1139
50. Wisniewski T, Frangione B (1996) Molecular biology of brain aging and neurodegenerative disorders. Acta Neurobiol Exp (Wars) 56(1): 267–279
51. Yagami T (2006) Cerebral arachidonate cascade in dementia: Alzheimer's disease and vascular dementia. Curr Neuropharmacol 4(1):87–100
52. Lin TN, Wang Q, Simonyi A, Chen JJ, Cheung WM, He YY, Xu J, Sun AY, Hsu CY, Sun GY (2004) Induction of secretory phospholipase A2 in reactive astrocytes in response to transient focal cerebral ischemia in the rat brain. J Neurochem 90(3):637–645
53. Moses GS, Jensen MD, Lue LF, Walker DG, Sun AY, Simonyi A, Sun GY (2006) Secretory PLA2-IIA: a new inflammatory factor for Alzheimer's disease. J Neuroinflammation 3:28
54. Bezprozvanny I, Mattson MP (2008) Neuronal calcium mishandling and the pathogenesis of Alzheimer's disease. Trends Neurosci 31(9): 454–463
55. Ueda K, Shinohara S, Yagami T, Asakura K, Kawasaki K (1997) Amyloid beta protein potentiates Ca^{2+} influx through L-type voltage-sensitive Ca^{2+} channels: a possible involvement of free radicals. J Neurochem 68(1):265–271
56. Mattson MP, Cheng B, Davis D, Bryant K, Lieberburg I, Rydel RE (1992) beta-Amyloid peptides destabilize calcium homeostasis and render human cortical neurons vulnerable to excitotoxicity. J Neurosci 12(2):376–389
57. Ueda K, Fukui Y, Kageyama H (1994) Amyloid beta protein-induced neuronal cell death: neurotoxic properties of aggregated amyloid beta protein. Brain Res 639(2):240–244
58. Yagami T, Ueda K, Asakura K, Nakazato H, Hata S, Kuroda T, Sakaeda T, Sakaguchi G, Itoh N, Hashimoto Y, Hori Y (2003) Human group IIA secretory phospholipase A2 potentiates Ca^{2+} influx through L-type voltage-sensitive Ca^{2+} channels in cultured rat cortical neurons. J Neurochem 85(3):749–758
59. Viola HM, Arthur PG, Hool LC (2007) Transient exposure to hydrogen peroxide causes an increase in mitochondria-derived superoxide as a result of sustained alteration in L-type Ca^{2+} channel function in the absence of apoptosis in ventricular myocytes. Circ Res 100(7):1036–1044
60. Nunomura A, Perry G, Aliev G, Hirai K, Takeda A, Balraj EK, Jones PK, Ghanbari H, Wataya T, Shimohama S, Chiba S, Atwood CS, Petersen RB, Smith MA (2001) Oxidative damage is the earliest event in Alzheimer disease. J Neuropathol Exp Neurol 60(8):759–767
61. Paola D, Domenicotti C, Nitti M, Vitali A, Borghi R, Cottalasso D, Zaccheo D, Odetti P, Strocchi P, Marinari UM, Tabaton M, Pronzato MA (2000) Oxidative stress induces increase in intracellular amyloid beta-protein production and selective activation of betaI and betaII PKCs in NT2 cells. Biochem Biophys Res Commun 268(2): 642–646
62. Harada J, Sugimoto M (1999) Activation of caspase-3 in beta-amyloid-induced apoptosis of cultured rat cortical neurons. Brain Res 842(2):311–323
63. Lee C, Park DW, Lee J, Lee TI, Kim YJ, Lee YS, Baek SH (2006) Secretory phospholipase A2 induces apoptosis through TNF-alpha and cytochrome *c*-mediated caspase cascade in murine macrophage RAW 264.7 cells. Eur J Pharmacol 536(1–2):47–53
64. Black SE (2005) Vascular dementia. Stroke risk and sequelae define therapeutic approaches. Postgrad Med 117(1):15–16, 19–25
65. in't Veld BA, Ruitenberg A, Hofman A, Stricker BH, Breteler MM (2001) Antihypertensive drugs and incidence of dementia: the Rotterdam Study. Neurobiol Aging 22(3):407–412
66. Schehr RS (1996) New treatments for acute stroke. Nat Biotechnol 14(11):1549–1554
67. Umemura K, Kawai H, Ishihara H, Nakashima M (1995) Inhibitory effect of clopidogrel, vapiprost and argatroban on the middle cerebral artery thrombosis in the rat. Jpn J Pharmacol 67(3):253–258
68. Hallenbeck JM (1994) Blood-damaged tissue interaction in experimental brain ischemia. Acta Neurochir Suppl (Wien) 60:233–237
69. Li Y, Sharov VG, Jiang N, Zaloga C, Sabbah HN, Chopp M (1995) Ultrastructural and light microscopic evidence of apoptosis after middle cerebral artery occlusion in the rat. Am J Pathol 146(5): 1045–1051
70. Yagami T, Ueda K, Asakura K, Hayasaki-Kajiwara Y, Nakazato H, Sakaeda T, Hata S, Kuroda T, Takasu N, Hori Y (2002) Group IB secretory phospholipase A2 induces neuronal cell death via apoptosis. J Neurochem 81(3):449–461
71. Yagami T, Ueda K, Asakura K, Sakaeda T, Hata S, Kuroda T, Sakaguchi G, Itoh N, Hashimoto Y, Hori Y (2003) Porcine pancreatic group IB secretory phospholipase A2 potentiates Ca^{2+} influx through L-type voltage-sensitive Ca^{2+} channels. Brain Res 960(1–2):71–80
72. Prat A, Antel J (2005) Pathogenesis of multiple sclerosis. Curr Opin Neurol 18(3):225–230
73. Pinto F, Brenner T, Dan P, Krimsky M, Yedgar S (2003) Extracellular phospholipase A2 inhibitors suppress central nervous system inflammation. Glia 44(3):275–282

74. Cunningham TJ, Yao L, Oetinger M, Cort L, Blankenhorn EP, Greenstein JI (2006) Secreted phospholipase A2 activity in experimental autoimmune encephalomyelitis and multiple sclerosis. J Neuroinflammation 3:26
75. Profyris C, Cheema SS, Zang D, Azari MF, Boyle K, Petratos S (2004) Degenerative and regenerative mechanisms governing spinal cord injury. Neurobiol Dis 15(3):415–436
76. Titsworth WL, Cheng X, Ke Y, Deng L, Burckardt KA, Pendleton C, Liu NK, Shao H, Cao QL, Xu XM (2009) Differential expression of sPLA2 following spinal cord injury and a functional role for sPLA2-IIA in mediating oligodendrocyte death. Glia 57(14):1521–1537
77. Yegin A, Akbas SH, Ozben T, Korgun DK (2002) Secretory phospholipase A2 and phospholipids in neural membranes in an experimental epilepsy model. Acta Neurol Scand 106(5):258–262
78. Visioli F, Rodriguez de Turco EB, Kreisman NR, Bazan NG (1994) Membrane lipid degradation is related to interictal cortical activity in a series of seizures. Metab Brain Dis 9(2):161–170
79. Lambeau G, Ancian P, Barhanin J, Lazdunski M (1994) Cloning and expression of a membrane receptor for secretory phospholipases A2. J Biol Chem 269(3):1575–1578
80. Lambeau G, Barhanin J, Schweitz H, Qar J, Lazdunski M (1989) Identification and properties of very high affinity brain membrane-binding sites for a neurotoxic phospholipase from the Taipan venom. J Biol Chem 264(19):11503–11510
81. Copic A, Vucemilo N, Gubensek F, Krizaj I (1999) Identification and purification of a novel receptor for secretory phospholipase A(2) in porcine cerebral cortex. J Biol Chem 274(37):26315–26320
82. Dev KK, Foged C, Andersen H, Honore T, Henley JM (1997) High-affinity binding sites for ^{125}I-labelled pancreatic secretory phospholipase A2 in rat brain. Brain Res Mol Brain Res 49(1–2):120–126
83. Higashino K, Ishizaki J, Kishino J, Ohara O, Arita H (1994) Structural comparison of phospholipase-A2-binding regions in phospholipase-A2 receptors from various mammals. Eur J Biochem 225(1):375–382
84. Nakajima M, Hanasaki K, Ueda M, Arita H (1992) Effect of pancreatic type phospholipase A2 on isolated porcine cerebral arteries via its specific binding sites. FEBS Lett 309(3):261–264
85. Kishino J, Ohara O, Nomura K, Kramer RM, Arita H (1994) Pancreatic-type phospholipase A2 induces group II phospholipase A2 expression and prostaglandin biosynthesis in rat mesangial cells. J Biol Chem 269(7):5092–5098
86. Arita H, Hanasaki K, Nakano T, Oka S, Teraoka H, Matsumoto K (1991) Novel proliferative effect of phospholipase A2 in Swiss 3T3 cells via specific binding site. J Biol Chem 266(29):19139–19141
87. Hanasaki K, Arita H (1992) Characterization of a high affinity binding site for pancreatic-type phospholipase A2 in the rat. Its cellular and tissue distribution. J Biol Chem 267(9):6414–6420
88. Ishizaki J, Hanasaki K, Higashino K, Kishino J, Kikuchi N, Ohara O, Arita H (1994) Molecular cloning of pancreatic group I phospholipase A2 receptor. J Biol Chem 269(8):5897–5904
89. Yokota Y, Hanasaki K, Ono T, Nakazato H, Kobayashi T, Arita H (1999) Suppression of murine endotoxic shock by sPLA2 inhibitor, indoxam, through group IIA sPLA2-independent mechanisms. Biochim Biophys Acta 1438(2):213–222
90. Kolko M, de Turco EB, Diemer NH, Bazan NG (2002) Secretory phospholipase A2-mediated neuronal cell death involves glutamate ionotropic receptors. Neuroreport 13(15):1963–1966
91. Ray SK, Banik NL (2003) Calpain and its involvement in the pathophysiology of CNS injuries and diseases: therapeutic potential of calpain inhibitors for prevention of neurodegeneration. Curr Drug Targets CNS Neurol Disord 2(3):173–189
92. Sorce S, Krause KH (2009) NOX enzymes in the central nervous system: from signaling to disease. Antioxid Redox Signal 11(10): 2481–2504
93. Bey EA, Xu B, Bhattacharjee A, Oldfield CM, Zhao X, Li Q, Subbulakshmi V, Feldman GM, Wientjes FB, Cathcart MK (2004) Protein kinase C delta is required for p47phox phosphorylation and translocation in activated human monocytes. J Immunol 173(9): 5730–5738
94. Simonyi A, He Y, Sheng W, Sun AY, Wood WG, Weisman GA, Sun GY (2010) Targeting NADPH oxidase and phospholipases A2 in Alzheimer's disease. Mol Neurobiol 41(2–3):73–86
95. Brennan AM, Suh SW, Won SJ, Narasimhan P, Kauppinen TM, Lee H, Edling Y, Chan PH, Swanson RA (2009) NADPH oxidase is the primary source of superoxide induced by NMDA receptor activation. Nat Neurosci 12(7):857–863
96. Shelat PB, Chalimoniuk M, Wang JH, Strosznajder JB, Lee JC, Sun AY, Simonyi A, Sun GY (2008) Amyloid beta peptide and NMDA induce ROS from NADPH oxidase and AA release from cytosolic phospholipase A2 in cortical neurons. J Neurochem 106(1):45–55
97. Sherry ST, Ward M, Sirotkin K (1999) dbSNP-database for single nucleotide polymorphisms and other classes of minor genetic variation. Genome Res 9(8):677–679
98. Eckert GP, Schaeffer EL, Schmitt A, Maras A, Gattaz WF (2011) Increased brain membrane fluidity in schizophrenia. Pharmacopsychiatry 44(4):161–162
99. Schaeffer EL, Gattaz WF, Eckert GP (2012) Alterations of brain membranes in Sshizophrenia: impact of phospholipase A2. Curr Top Med Chem
100. Barbosa NR, Junqueira RM, Vallada HP, Gattaz WF (2007) Association between BanI genotype and increased phospholipase A2 activity in schizophrenia. Eur Arch Psychiatry Clin Neurosci 257(6): 340–343
101. Bate C, Reid S, Williams A (2004) Phospholipase A2 inhibitors or platelet-activating factor antagonists prevent prion replication. J Biol Chem 279(35):36405–36411
102. Bate C, Tayebi M, Williams A (2008) Sequestration of free cholesterol in cell membranes by prions correlates with cytoplasmic phospholipase A2 activation. BMC Biol 6:8
103. Yoshino H, Tomiyama H, Tachibana N, Ogaki K, Li Y, Funayama M, Hashimoto T, Takashima S, Hattori N (2010) Phenotypic spectrum of patients with PLA2G6 mutation and PARK14-linked parkinsonism. Neurology 75(15):1356–1361
104. Klivenyi P, Beal MF, Ferrante RJ, Andreassen OA, Wermer M, Chin MR, Bonventre JV (1998) Mice deficient in group IV cytosolic phospholipase A2 are resistant to MPTP neurotoxicity. J Neurochem 71(6):2634–2637
105. Tariq M, Khan HA, Al Moutaery K, Al Deeb S (2001) Protective effect of quinacrine on striatal dopamine levels in 6-OHDA and MPTP models of Parkinsonism in rodents. Brain Res Bull 54(1):77–82
106. Pereira MP, Hurtado O, Cardenas A, Bosca L, Castillo J, Davalos A, Vivancos J, Serena J, Lorenzo P, Lizasoain I, Moro MA (2006) Rosiglitazone and 15-deoxy-Delta12,14-prostaglandin J2 cause potent neuroprotection after experimental stroke through noncompletely overlapping mechanisms. J Cereb Blood Flow Metab 26(2):218–229
107. Yagami T, Ueda K, Asakura K, Takasu N, Sakaeda T, Itoh N, Sakaguchi G, Kishino J, Nakazato H, Katsuyama Y, Nagasaki T, Okamura N, Hori Y, Hanasaki K, Arimura A, Fujimoto M (2003) Novel binding sites of 15-deoxy-Delta12,14-prostaglandin J2 in plasma membranes from primary rat cortical neurons. Exp Cell Res 291(1):212–227