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Review

Beyond PDE4 inhibition: A comprehensive review on downstream cAMP signaling in the central nervous system

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ABSTRACT

Cyclic adenosine monophosphate (cAMP) is a key second messenger that regulates signal transduction pathways pivotal for numerous biological functions. Intracellular cAMP levels are spatiotemporally regulated by their hydrolyzing enzymes called phosphodiesterases (PDEs). It has been shown that increased cAMP levels in the central nervous system (CNS) promote neuroplasticity, neurotransmission, neuronal survival, and myelination while suppressing neuroinflammation. Thus, elevating cAMP levels through PDE inhibition provides a therapeutic approach for multiple CNS disorders, including multiple sclerosis, stroke, spinal cord injury, amyotrophic lateral sclerosis, traumatic brain injury, and Alzheimer's disease. In particular, inhibition of the cAMP-specific PDE4 subfamily is widely studied because of its high expression in the CNS. So far, the clinical translation of full PDE4 inhibitors has been hampered because of dose-limiting side effects. Hence, focusing on signaling cascades downstream activated upon PDE4 inhibition presents a promising strategy, offering novel and pharmacologically safe targets for treating CNS disorders. Yet, the underlying downstream signaling pathways activated upon PDE(4) inhibition remain partially elusive. This review provides a comprehensive overview of the existing knowledge regarding downstream mediators of cAMP signaling induced by PDE4 inhibition or cAMP stimulators. Furthermore, we highlight existing gaps and future perspectives that may incentivize additional downstream research concerning PDE(4) inhibition, thereby providing novel therapeutic approaches for CNS disorders.

1. Introduction

The nucleotides 3′-5′-cyclic adenosine monophosphate (cAMP) and 3′-5′-cyclic guanosine monophosphate (cGMP) function as key second messengers in various intracellular signaling pathways. Second messengers are small, rapidly diffusing molecules that amplify and transduce extracellular signals, which initiate a cascade of signal transduction pathways, leading to various physiological cellular effects [1,2]. cAMP, first identified in 1985, is of particular interest since it is the key regulator of numerous biological processes across all body tissues, including but not limited to immune function, metabolism, and neurotransmission [3]. Moreover, cAMP regulates every aspect of the cellular life cycle, including proliferation, migration, differentiation, and survival [4,5]. A family of enzymes called adenylyl cyclases (ACs) produce cAMP by converting adenosine 5'-triphosphate (ATP) (Fig. 1). ACs are predominantly activated through ligand binding of transmembrane G protein-coupled receptors (GPCRs), as thoroughly reviewed by Devasani and Yao [6]. GPCRs undergo a conformational change upon ligand binding, leading to dissociation of the G-protein, a heterotrimer consisting of α, β, and γ subunits. Subsequently, the subunits Gα and Gβγ independently activate (e.g., Gαs) or inhibit (e.g., Gαi/o) ACs [6]. Additionally, the enzymatic activity of ACs can be

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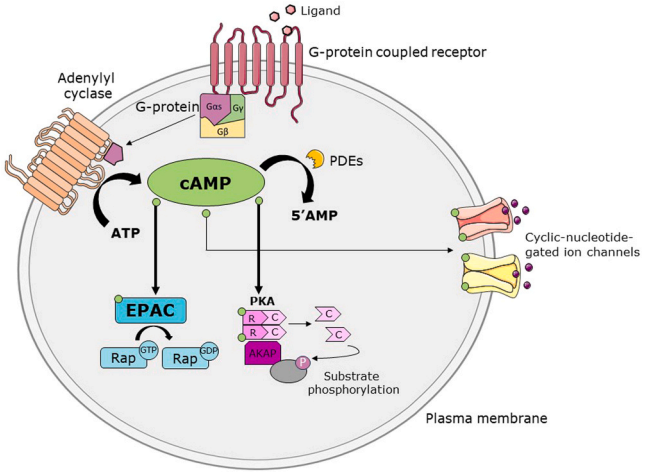

Fig. 1. : Synthesis, degradation, and main downstream effectors of cAMP. The binding of a ligand to G-protein coupled receptors with a Gαs subunit results in the activation of adenylyl cyclase. Adenylyl cyclase converts ATP to cAMP, whereas PDEs hydrolyze cAMP to 5'AMP. cAMP activates PKA, EPAC, and cyclic-nucleotide-gated ion channels. The binding of cAMP to the R units in the heterotetrameric PKA results in the dissociation of the C units and, hence, activation of PKA. Eventually, PKA mediates its biological effects by phosphorylation of downstream substrates, to which it is clustered through AKAPs. cAMP-mediated activation of EPAC leads to the activation of Rap GTPases. Cyclic-nucleotide-gated ion channels can be directly activated by cAMP, resulting in altered gating properties of these ion channels. Notably, this is a simplified representation as it does not show subcellular localization of cAMP domains. *cAMP, Cyclic adenosine monophosphate; 5'AMP, 5'Adenosine monophosphate; PDEs, Phosphodiesterases; PKA, Protein kinase A; AKAP, A-kinase anchoring protein. R unit, Regulatory unit; C unit, catalytic unit; EPAC, Exchange protein activated by cAMP; Rap, Ras-associated protein; GTP, Guanosine triphosphate.*

modulated intracellularly by protein kinases or calcium fluctuations [5,7]. Therefore, cAMP signaling can be activated by multiple stimuli through ACs, which in turn are regulated by various feedback loops. Furthermore, cAMP signaling is spatiotemporally regulated and catabolized by phosphodiesterases (PDEs), a family of enzymes that hydrolyze cAMP and/or cGMP to 5' AMP or 5' GMP, respectively. The PDE enzyme superfamily consists of 11 gene families (PDE1–11) encoded by 21 genes (e.g., PDE4A-D), each one coding for different isoforms (e.g., PDE4D1–9). This results in a total of at least 77 different protein-coding isoforms that differ in substrate specificity, tissue expression profile, and subcellular localization [8,9]. PDE4, PDE7, and PDE8 are selective for cAMP breakdown, whereas PDE5, PDE6, and PDE9 specifically hydrolyze cGMP. The remaining families exhibit dual specificity, with a varying affinity for the cyclic nucleotides depending on the isoform [8,10,11]. Therefore, influencing PDE activity allows the control of cAMP levels in a cell type-specific and spatiotemporal manner.

cAMP has been shown to exert beneficial effects on various neurological processes in the central nervous system (CNS), including modulating memory processes, suppressing neuroinflammation, promoting repair processes by boosting neuroplasticity and remyelination, stimulating neurotransmission and cognition [12–17]. Because of its crucial role in these CNS processes, cAMP is a promising target for neurodegenerative disorders. Unfortunately, raising cAMP through exogenous administration of cAMP is associated with dose-limiting toxicities, such as hypercalcemia [18]. Consequently, elevating cAMP levels intracellularly by inhibiting PDEs offers a therapeutic strategy for Alzheimer's disease (AD), multiple sclerosis (MS), schizophrenia, traumatic brain injury (TBI), and other diseases [8,19,20]. PDE4, in particular, has been extensively studied as a target for CNS disorders, as reviewed by *Blokland et al.* [20]. Although PDE4 inhibitors are used in the clinic to treat chronic obstructive pulmonary disease and psoriasis, their clinical application for CNS indications is hampered due to dose-dependent side effects, including emesis, nausea, and diarrhea, linked to the therapeutic effective dose. Yet, ibudilast, roflumilast, and zatolmilast are in clinical trials for various CNS diseases, such as AD and fragile X syndrome [21–23]. Thus, cAMP modulation in the CNS through PDE4 inhibition remains clinically relevant. Approved PDE4 inhibitors and the ones in clinical trials are thoroughly reviewed by Crocetti, Floresta [24] and Li, He [25]. In the past decade, research in the PDE field, including ours, has focused on selective inhibition of PDE4 subtypes (e.g., PDE4D) or isoforms (e.g., PDE4D4) [26,27]. Still, a significant knowledge gap concerns the actual effect of PDE inhibition on the downstream cAMP-mediated signaling pathways, associated effectors, and interactive effects leading to complex physiological cell changes that improve CNS disease outcomes [9,20].

cAMP has three main downstream effectors: 1) protein kinase A (PKA), 2) exchange protein activated by cAMP (EPAC), and 3) ion channels, i.e., cyclic-nucleotide-gated ion channels (CNGCs) and hyperpolarization-activated nucleotide-gated (HCN) channels [2,3] (Fig. 1). Among these three, cAMP-dependent PKA, a heterotetrameric enzyme comprising two regulatory (R) and two catalytic (C) subunits, is the most extensively studied. Specifically, PKA activation occurs through cAMP binding to the R units, inducing dissociation and, thus, activation of the C subunits. Subsequently, the liberated C subunits phosphorylate serine and threonine amino acid residues on target substrates, thereby initiating numerous signaling pathways. The transcription factor cAMP-response binding protein (CREB) is the most well-known target of PKA. Upon phosphorylation, CREB translocates to the nucleus, where it binds to cAMP-response elements (CREs) in the promotor region of various target genes, thereby regulating their expression. The cAMP-PKA-CREB pathway has been described to mediate multiple CNS functions, including neuronal survival, neuroplasticity underlying learning and memory, differentiation of oligodendrocyte precursor cells (OPCs), and neuroinflammation [4,28–30]. Although CREB might seem like a promising target in CNS diseases, it is non-specific since cAMP/CREB is ubiquitously expressed in all body tissues [28]. Other cAMP-responsive transcription factors regulated by PKA include nuclear factor-κB (NF-κB), activating transcription factor-1/2 (ATF-1/2) [31]. In addition, PKA phosphorylates ion channels, motor proteins, and other enzymes such as protein kinase C (PKC), phosphoinositide 3-kinase (PI3K), phospholipase C (PLC), and members of the mitogen-activated protein kinase (MAPK) family (i.e., extracellular signal-regulated kinase (ERK), c-Jun N-terminal kinase (JNK), and p38 MAPK) [2,4,32]. Additionally, the specificity of cAMP/PKA signal transduction is mediated by A-kinase anchoring proteins (AKAPs) since they locate PKA closer to its substrates and control its subcellular localization. This facilitates efficient spatial and temporal regulation of cAMP/PKA signaling [2,33]. EPAC, a guanine nucleotide exchange factor (GEF), is cAMP's second main downstream target. Upon activation, EPAC regulates kinases, including Raf and ERK, and activates the small GTPases Ras-associated protein (Rap)1 and Rap2 through guanine-nucleotide exchange. These factors control various processes, including cell adhesion, cell-cell junction formation, exocytosis, and cellular functions such as proliferation, differentiation, and migration [10,11,34]. In the CNS, EPAC plays a role in neuronal differentiation, particularly in neuronal polarity and cone movement, and microglia-mediated neuroinflammation [10,35]. Thirdly, cAMP also binds to and directly activates CNGCs, which are voltage-dependent non-selective cation channels found in many tissues, including the heart, kidney, and brain [11]. HCN channels are typically associated with neuronal and cardiac excitability by conducting calcium, sodium, and potassium. Additionally, cAMP has been found to regulate the length of myelin sheaths, vital for axonal signal conduction and proper CNS functioning, through HCN2 channels [36,37]. Since the role of CNGCs in both the healthy and diseased CNS remains poorly understood, they fall beyond the scope of this review, which primarily focuses on cAMP/PKA and cAMP/EPAC signaling.

This review summarizes the downstream effects of cAMP elevations upon PDE4 inhibition and cAMP analogs or activators. While PDE4 inhibitors offer a more cell-type specific approach and are clinically relevant, cAMP analogs or activators can potentially elicit similar downstream effects compared to PDE4 inhibitors, aiding further investigation for novel downstream targets. Therefore, we provide a comprehensive overview of cAMP signaling and downstream effectors in the CNS while focusing on the following key CNS processes: (1) neuroinflammation, (2) neuroplasticity, (3) neurotransmission and long-term potentiation (LTP), (4) neuronal survival, and (5) myelination. The most commonly used PDE4 modulators, cAMP analogs, and modulators of cAMP-related signaling molecules in the discussed studies are summarized in Table A.1. Although these specific modulators are not explicitly mentioned in the test, the table provides a valuable overview for those interested in this information. In addition, implications in disease and existing literature regarding other PDE inhibitors will be briefly discussed. Lastly, future directives are outlined that could help to unravel downstream cAMP signaling to provide more specific targets to treat CNS-related diseases.

2. Neuroinflammation

Resident CNS immune cells, including microglia and astrocytes, as well as infiltrating inflammatory cells, contribute to neuroinflammatory responses in neurodegenerative disorders such as AD, Parkinson's disease (PD), spinal cord injury (SCI), TBI, stroke, and amyotrophic lateral sclerosis (ALS) [38–40]. In homeostatic conditions, these inflammatory cells remove harmful substances and tissue debris, promote CNS repair, and stimulate neuroplasticity [41,42]. In contrast, an exacerbated and chronic inflammatory response is often observed during neurodegenerative diseases, resulting in increased neuronal death, demyelination, attenuated CNS repair, and escalated tissue damage [43,44]. Consequently, previous studies have focused on modulating neuroinflammatory responses in CNS disorders. cAMP regulation through PDE inhibitors or cAMP analogs has already been shown to be a promising therapeutic strategy to orchestrate inflammatory responses [4,31]. Here, we will focus on the cAMP pathways involved in microglial and astrocyte modulation.

2.1. cAMP signaling upon PDE4 inhibition in neuroinflammation

Accumulating evidence suggests the importance of PDEs, especially PDE4B, in regulating inflammatory responses of microglia and astrocytes. *Pde4b* was found to be acutely upregulated in microglia following traumatic SCI in rodents. Additionally, stimulation with pro-inflammatory cytokines tumor necrotic factor α (TNF-α) and interleukin 1β (IL-1β) upregulated *Pde4b* in rodent-derived astrocytes and microglia [45–47]. Interestingly, the ablation of *Pde4b* reduced TNF-α and IL-1β expression upon lipopolysaccharide (LPS)-induced inflammation on primary rat microglia [48]. Thus, these results suggest that inflammation regulates PDE4B and, conversely, that PDE4B orchestrates inflammatory responses. The interplay between inflammation and PDE4 is suggested through the NF-κB pathway, which is well known to promote the production of inflammatory cytokines, including TNF-α, IL-6, IL-1β, and inducible nitric oxide synthase (iNOS) in glial cells [49]. Research showed that inhibition of NF-κB translocation to the nucleus prevented TNF-α-mediated upregulation of *Pde4* expression and cAMP decreases in murine microglia, suggesting a link between the NF-κB pathway and *Pde4* expression [50]. Conversely, PDE4 inhibition attenuates the NF-κB pathway in microglia and astrocytes. TNF-α downregulates cAMP, which is abolished in the murine microglia cell line upon inhibition of PDE4, PKA, MEK, and NF-κB translocation. These results imply that downstream kinases PKA and MEK are involved in cAMP-mediated NF-κB regulation. The inhibition of PKA can lead to reduced PKA-mediated phosphorylation of upstream molecules in the NF-κB signaling pathway, like p50, resulting in increased inflammation. On the other hand, PKA itself can promote inflammation, postulated through p65 phosphorylation, a dimer of NF-κB, resulting in NF-κB translocation to the nucleus [51,52]. However, PDE4 inhibits the translocation of phosphorylated p65 to the nucleus in a TNF-α stimulated murine microglia cell line, leading to a reduced inflammatory response [50]. Other mechanisms of PDE4 inhibition-mediated anti-inflammatory effects on glial cells are reducing IκB degradation and inhibiting IKK activation, thereby keeping NF-κB proteins sequestered in the cytoplasm [45,53,54].

Besides the NF-κB pathway, other pathways are pivotal in regulating glial inflammation. PDE4 inhibitor roflupram increased AMP-activated protein kinase (AMPK) phosphorylation and elevated the protein expression of the histone/protein deacetylase SIRT1, a molecule downstream of AMPK [55]. Furthermore, inhibition of AMPK or SIRT1 abolished the anti-inflammatory effects of PDE4 inhibition on LPS-stimulated BV2 microglial cells, indicating that PDE4 inhibition attenuates inflammation through the AMPK/SIRT1 pathway [55]. The beneficial effects of SIRT1 on microglia are postulated through stimulating repair-promoting microglia, decreasing inflammasome activation, and inhibiting the NF-κB pathway [56,57]. In contrast, SIRT1 activation promotes an inflammatory astrocyte phenotype [57]. However, the direct result of the interaction between PDE inhibition and SIRT1 in astrocytes remains to be elucidated. CREB has also been shown to play an essential role in reducing the production of pro-inflammatory cytokines (TNF-α, IL-1β, and IL-6) upon PDE4 inhibition in a murine microglia cell line. These anti-inflammatory effects were nullified upon PKA inhibition, suggesting a PKA/CREB-mediated signaling pathway for suppressing microglial inflammation [58]. Neuroinflammation can also be modulated through inhibition of MAPK signaling, as demonstrated with other PDE inhibitors [59]. Yet, the exact role of this pathway upon PDE4 inhibition is not known.

2.2. cAMP signaling upon cAMP stimulants or cAMP analogs in neuroinflammation

The role of PKA signaling in modulating inflammation became evident when cAMP analogs were used. Incubation of primary rat microglia with the PKA-stimulating cAMP analog 6-Bnz-cAMP followed by LPS stimulation decreased TNF-α and IL-1β levels and increased the levels of anti-inflammatory cytokine IL-10. Furthermore, PKA inhibition abolished these beneficial effects, implying that the reduction in pro-inflammatory cytokine release is mediated through PKA. Downstream modulators in orchestrating cytokine secretion upon PKA signaling are p38 and glycogen synthase kinase-3 β (GSK3β) since PKA activation decreased p38 phosphorylation and enhanced GSK3β phosphorylation. Importantly, as opposed to other kinases, GSK3β is inactivated by phosphorylation. Additionally, the EPAC-specific cAMP analog 8-CPT-cAMP) attenuated TNF-α but not IL-1β and IL-10 levels in LPS-stimulated rat microglia. Interestingly, EPAC stimulation also promoted GSK-3β phosphorylation similar to PKA activation [13]. Consequently, it is hypothesized that PKA and EPAC activation attenuates pro-inflammatory cytokine release by inhibiting the p38 MAPK pathway and GSK3β signaling. *Ghosh et al.* indicated that PKA inhibition reduced the inflammatory markers iNOS and cyclooxygenase 2 (COX2) in TNF-α-stimulated microglia [50]. This implies that different downstream effectors of PKA regulate the release of specific cytokines. This hypothesis is supported by a study showing that PKA and CREB inhibition abolished anti-inflammatory cytokines Arg1, CD206, and IL-4 upon stimulation of the Gα-coupled GPCR GLP-1R. These results suggest a role for PKA/CREB in mediating an anti-inflammatory phenotype upon GPCR signaling. Moreover, p38 inhibition suppressed *Arg1* and *Cd206* expression, while *Il-4* expression was inhibited by JNK inhibition. Therefore, *Arg1* and *Cd206* expression was hypothesized to be regulated through PKA/p38/CREB signaling, whereas *Il-4* expression is suggested to be controlled by PKA/JNK/CREB signaling [60]. PKA-stimulating cAMP analogs, but not the EPAC-selective analogs, could increase the

expression of *Arg1* in an IL-4-stimulated murine microglia cell line [61]. Hence, PKA and EPAC are suggested to orchestrate inflammatory responses through distinct pathways that may overlap.

Lastly, regulating the microglial and astrocyte phenotype is key to determining their function. For instance, cytoskeleton arrangements are demanded to mediate phagocytosis of cellular debris. Interestingly, cAMP signaling also modulates cytoskeleton adaptations of microglia and astrocytes during inflammation. Research indicated that PKA inhibition or stimulation in microglial cells diminishes or increases their phagocytic capacity, respectively [50,61]. EPAC has been demonstrated to remodel the cytoskeleton by reducing a hypertrophic, inflammatory astrocyte profile and attenuating the round inflammatory microglia phenotype in SCI, possibly through the STAT3 signaling pathway [62]. PKA also regulates glutamate uptake in rat glial cells by influencing actin polymerization. Specifically, PKA inhibition enhanced glutamate uptake [63]. The uptake of neurotransmitters such as glutamate is a critical function of glial cells to protect the CNS environment and thereby reduce neuroinflammation, pointing to the importance of PKA regulation.

2.3. Summary

cAMP pathway modulation can be leveraged to mediate neuroinflammatory responses in CNS disorders. PDE4 inhibition decreases neuroinflammation by attenuating the NF-κB pathway and boosting AMPK/SIRT and PKA/CREB signaling in microglia and astrocytes. cAMP analogs have been shown to promote anti-inflammatory responses by inhibiting the p38 MAPK and GSK3β signaling pathways, mediated by both EPAC and PKA (Fig. 2).

3. Neuroplasticity

During life, different types of neuroplasticity impact brain function and structure [64,65]. First, structural neuroplasticity refers to neurogenesis and the modulation of the physical appearance of existing [66–68]. Second, the growth of axons and the development of dendrites into axons are essential for the polarizing plasticity of neurons [69,70]. Third, functional neuroplasticity, also called synaptic plasticity, refers to the modification of the strength or efficacy of transmission of preexisting synapses in an activity-dependent manner [71]. The strengthening of synapses represents the cellular foundation underlying learning and memory, cognition, and behavior. Neurodegenerative diseases such as AD, PD, or CNS injury, including SCI or stroke, are accompanied by impaired neuroplasticity, often resulting in cognitive and behavioral problems [72–75]. Therefore, interventions designed to enhance neuroplasticity are predicted to improve disease symptoms or slow disease progression. cAMP and its downstream effectors are essential in regulating neuroplasticity. In this part, we will review current evidence on the effect of cAMP elevation through PDE inhibition and cAMP stimulants and analogs on downstream signaling within neuroplastic processes.

3.1. cAMP signaling upon PDE4 inhibition in neuroplasticity

There is accumulating evidence that inhibition of PDE4 alters structural and functional neuroplasticity in health and disease. Chronic PDE4 inhibition through oral administration of roflumilast or rolipram in the APP/PS1 mouse model for AD increased levels of phosphorylated CREB (pCREB) and brain-derived neurotrophic factor (BDNF) [76]. pCREB is suggested to modulate neuroplasticity by promoting the gene

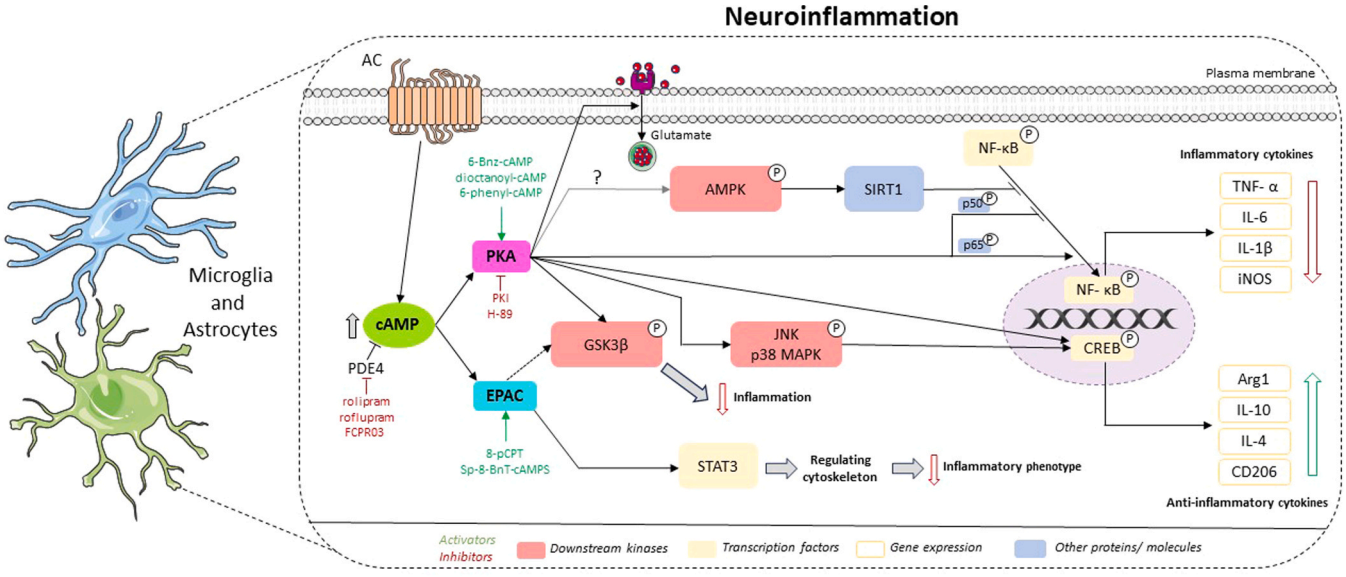

Fig. 2. : Downstream signaling upon cAMP stimulation or PDE4 inhibition in neuroinflammatory responses**.** Promoting cAMP signaling through the inhibition of PDE4 (rolipram, roflupram, or FCPR03) or cAMP analogs specific for PKA (6-Bnz-cAMP, dioctanoyl-cAMP, or 6-phenyl-cAMP) and/or EPAC (8-pCPT or Sp-8-BnT-cAMPS) can influence the inflammatory outcomes in microglia and astrocytes. PDE4 inhibition attenuates neuroinflammation via PKA/AMPK/SIRT1 signaling and PKA-induced phosphorylating of p50, leading to reduced translocation of the inflammation-stimulating transcription factor NF-κB and hence, decreased production of inflammatory products (TNF-α, IL-6, IL-1B, and iNOS). On the other hand, the phosphorylation of p65 by PKA can promote inflammation via NF-κB translocation. PDE4 inhibition leads to PKA-mediated CREB phosphorylation directly or indirectly through JNK and p38 MAPK, which promotes the expression of anti-inflammatory genes (Arg1, IL-10, IL-4, and CD206). The removal of glutamate during inflammation can be mediated through PKA signaling. cAMP analogs for PKA and EPAC exert anti-inflammatory effects through GSK3β phosphorylation. In addition, EPAC promotes STAT3 signaling, which has been shown to modulate the glial cytoskeleton towards an anti-inflammatory cell phenotype. *AC, Adenylyl cyclase; PDE4, Phosphodiesterase 4; cAMP, Cyclic adenosine monophosphate; PKA, Protein kinase A; 6-Bnz-cAMP, Rp-cAMPS-6-Bnz-cAMP; PKI, Protein kinase inhibitor; AMPK, Adenosine monophosphate-activated protein kinase; SIRT1, Silent information regulator 1; NF-κB, Nuclear factor κ B; CREB, cAMP-response element binding protein; JNK, c-Jun N-terminal kinase; MAPK, Mitogen-activated protein kinase; EPAC, Exchange protein directly activated by cAMP; 8-pCPT, 8-(4-chlorophenylthion)-2′-O-methyl-cyclic AMP; Sp-8-BnT-cAMPS, 8- Benzylthioadenosine- 3′, 5′- cyclic mono phosphorothioate; GSK3β, Glycogen synthase kinase-3 β; STAT3, Signal transducer and activator of transcription 3; TNF-α, Tumor necrotic factor a; IL-6, Interleukin 6; iNOS, Inducible Nitrogen oxide synthase; Arg1, Arginase 1.*

expression of growth factors (e.g., BDNF) and genes involved in neuronal survival (e.g., Bcl-2) [77]. Several studies showed that neuroplasticity is promoted by BDNF and subsequent tropomyosin receptor kinase B (TrkB) receptor activation [15,78]. TrkB receptor stimulation results in the activation of multiple signaling pathways involved in neurite outgrowth and synaptic plasticity, including the Ras/ERK pathway and the PI3K/AKT pathway [79]. Various studies used PKA and EPAC activators and inhibitors to elucidate whether neuroplastic processes are dependent or independent of PKA and EPAC. In rat dorsal root ganglion (DRG) neurons, PKA activation is not essential to induce neurite outgrowth [15]. In contrast, EPAC does appear to be involved in neuronal elongation and modulation of synaptic transmission as well as plasticity in rat DRG neurons [80–82]. The EPAC agonist 8-CPT-cAMP triggered neurite outgrowth in murine neurons, while using an inactive version of 8-CPT-cAMP omitted the effect, providing evidence of the involvement of EPAC in neurite growth [83]. Double inhibition of PKA and EPAC had a detrimental effect on neurite outgrowth. Oppositely, a weak outgrowth stimulating effect was observed upon PKA inhibition. Therefore, it is postulated that cAMP-induced CREB phosphorylation leading to neurite outgrowth is primarily mediated by EPAC [15]. Another study has shown that rolipram treatment of an *in vitro* co-culture of myelinated neurons enhanced neurite density and increased neurite outgrowth in a PKA-independent manner [84]. Additionally, the distinct conformational states of PDE4 that exhibit different sensitivities to rolipram inhibition were studied: the high-affinity (HARBS) and low-affinity rolipram binding state (LARBS). The PDE4 inhibitor rolipram has two enantiomers, R- and S-rolipram, which selectively inhibit the PDE4 HARBS conformer and PDE4 LARBS conformer, respectively. This study showed that inhibition of the HARBS by R-rolipram, rather than the inhibition of LARBS by S-rolipram, promoted neurite outgrowth. As tested using PKA inhibitors, these effects were mediated through the activation of EPAC and not through PKA [84]. However, PKA inhibition decreased CREB phosphorylation triggered by the general PDE inhibitor IBMX but not after stimulation with AC activator forskolin. These results suggest that high levels of cAMP caused by forskolin, unlike IBMX, which is expected to increase cAMP levels moderately, might be required to drive PKA-independent CREB phosphorylation [15].

Hasan et al. showed that chronic intracerebroventricular PDE4 inhibition by roflumilast improved spatial memory performance by increasing BDNF levels while decreasing inflammatory processes (*NFκβ* expression) in the hippocampus of an AD rat model [85]. Roflumilast treatment inhibited NFκβ/BACE1-mediated Aβ production in the hippocampus. Furthermore, oral administration of roflumilast exerted neuroprotective effects in a rat model for PD. These neuroprotective effects were mediated via CREB/BDNF/TrkB, which activates the PI3K/AKT pathway [86]. In addition, reduced levels of activated Rho and phosphomyosin were observed following the PDE4 inhibition, which points to Rho and phosphomyosin being responsible for the enhanced neurite outgrowth [84,87]. Moreover, PDE4 inhibition by rolipram increased PKA-mediated phosphorylation of vasodilator-stimulated phosphoprotein (VASP), a protein that is associated with the cytoskeleton and could be involved in neurite elongation [88,89].

Notably, not every type of PDE4 inhibitor causes increases in pCREB and subsequent elevation of BDNF levels. Despite improving cognition, chronic subcutaneous administration of the PDE4D inhibitor Gebr7a to APP/PS1 mice did not alter hippocampal pCREB, BDNF, synaptophysin, and PSD95 levels [90]. A derivative compound of Gebr7b, Gebr32a, improved spatial memory performance in another AD mouse model [27]. Moreover, we have shown that Gebr32a improved the neurite outgrowth of a mouse hippocampal cell line, even upon Aβ-induced reduction of neurite length. Gebr32a treatment did increase pCREB levels *in vitro* in mouse N2a cells, but the effect on hippocampal CREB phosphorylation remains to be studied [91]. We concurringly see BDNF emerging as a primary effector of plasticity processes. Yet, regarding other neurotrophic factors such as NGF and GDNF, little is known in the context of phosphodiesterase inhibition, cAMP signaling, and neuroplasticity. It was shown that the general PDE inhibitor ibudilast promotes GDNF expression in a model of PD and promotes NGF and GDNF expression in activated microglia [92,93]. It remains to be resolved whether the PDE(4) inhibition-induced increase in NGF and GDNF expression affects signaling-induced neuroplasticity.

An additional cAMP-mediated mechanism for neurite and axon growth is preventing growth cone collapse. cAMP induces a reversible internalization of Nogo 66 receptor 1 (NgR1), a receptor for myelin inhibitors [94,95]. NgR1 activation causes growth cone collapse and, therefore, inhibits neuronal plasticity in a Rho/ROCK-dependent manner [94,96]. A study showed that rolipram treatment decreases the cell surface expression of NgR1 in neuronal cells (NS-1) through NgR1 internalization. PKA stimulation did not induce internalization of NgR1, while EPAC activation did. Moreover, PKA inhibition did not block the NgR1 internalization induced by cAMP analog dibutyryl-cAMP (db-cAMP), whereas EPAC inhibition did [94]. This data suggests that cAMP-induced NgR1 internalization depends on EPAC rather than PKA activation. However, both EPAC and PKA activation through rolipram can inhibit the Rho pathway independent of NgR1 internalization, possibly preventing growth cone collapse [84,97,98].

cAMP-mediated functional plasticity (e.g., synaptic plasticity) involves AMPA and NMDA receptor activation in excitatory neurons. The GluA subunits of AMPA receptors are crucial in synaptic plasticity and memory formation. Synapse strengthening involves the trafficking of GluA1/GluA2 heterodimers to the synaptic membrane [99,100]. These receptors are subsequently replaced by the constitutive trafficking GluA2/GluA3 of heterodimers, maintaining long-lasting synaptic strengthening [101]. Upon PDE4 inhibition by rolipram, membrane surface expression of GluA1-AMPA receptors was increased in the hippocampus, suggested by increased mobilization and trafficking of resident synaptic endosomes [14]. After 1 hour of rolipram treatment, transcription of GluA1, potentially CREB-mediated, and translation of local pools of GluA1 mRNA occurs, increasing the total GluA1-AMPA receptor expression [14]. Moreover, it was shown that BDNF regulates this local translation of GluA1. Using CaMKII, the incorporation of GluA1-AMPA receptors into the postsynaptic membrane is even more enhanced, possibly through CaMKII-mediated AC activation [102].

The application of PDE4 inhibitors shows excellent therapeutic potential for neurodegeneration and neurotrauma. However, therapeutic PDE4 inhibition in the CNS coincides with emetic adverse effects. A promising approach to avoiding these side effects while still enhancing neuroplastic processes is the selective inhibition of PDE4 genes and isoforms. Selective targeting of the PDE4B or PDE4D subtypes already circumvented the emetic side effects [26]. A mouse model study with disrupted *Pde4b1* showed increased pCREB and ERK1/2 phosphorylation levels, enhanced hippocampal neurogenesis, and increased baseline synaptic transmission. Together, these data suggest a role for PDE4B in regulating synaptic plasticity [103]. Moreover, we have shown that several PDE4D isoforms are aberrantly expressed in the temporal lobe of AD patients [104]. Knocking down long PDE4D isoforms (PDE4D3-D5, PDE4D7-D9) enhanced the neurite outgrowth of HT-22 cells. In contrast, this effect was not observed when the expression of short PDE4D isoforms (PDE4D1-D2, PDE4D6) was suppressed. PDE4D long isoforms are tethered in particular subcellular areas and anchored to AKAPs and scaffolding proteins, probably enabling them to exert specific functions in neurite outgrowth. For example, it was shown that the PDE4D3/-mAKAP complex tightly regulates axonal growth in neurons [105]. On the other hand, PDE4D5 preferably binds to B-arrestin as a scaffolding protein and is therefore located near several GPCRs. PDE4D9 can bind to and regulate the function of β2-adrenergic receptors, of which the activation promotes spine generation in APP/PS1 mice [106,107]. Ultimately, the effects of individual PDE4D isoforms on neuroplasticity and their association with particular complexes emphasize the intricate regulatory mechanisms within the cAMP signaling pathway. Whether

different PDE4D isoforms and isoform complexes initiate distinguished signaling cascades is likely but remains to be elucidated.

3.2. cAMP signaling upon cAMP stimulants or cAMP analogs in neuroplasticity

Some hydrolyzed derivatives of cAMP, such as db-cAMP and 8-bromo-cAMP (8-Br-cAMP), as well as the AC stimulator forskolin, have been described as neurite outgrowth stimulating agents in several neuronal cell types [108–110]. A study using the human neuroblastoma (SH-SY5Y) cell line treated with db-cAMP enhanced neurite formation and resulted in GAP43, neuronal tubulin, tau, and MAP1B accumulation in the cell, referred to as neuronal growth and plasticity proteins. PKA inhibition and concurrent db-cAMP administration inhibited neurite elongation. In contrast, adding a MEK/ERK or p38 inhibitor showed no effect on db-cAMP-mediated neurite elongation. This suggests that PKA is essential for initial SH-SY5Y cell differentiation upon db-cAMP. However, when PKA was inhibited after 48 h of db-cAMP stimulation, the db-cAMP-induced neurite elongation was not affected, indicating that PKA is not required in this later stage. In line with previous results, inhibition of MEK/ERK did not affect neurite outgrowth, while inhibition of PI3K induced neurite retraction. Consequently, PKA and PI3K seem essential in this elongation model, while ERK activation is not required [109]. Another study showed that db-cAMP upregulated arginase 1 (Arg1) expression and increased polyamine synthesis in cerebellar rat neurons, promoting neurite growth [111].

Furthermore, it was shown that forskolin, db-cAMP, and 8-Br-cAMP caused significant increases in neurite length in rat NS-1 cells. However, co-treatment with PKA inhibitors did not block forskolin-elicited neurite extension. This data suggests that the cAMP-induced neurite outgrowth in NS-1 cells is PKA-independent [112]. Another study using a striatal neuronal cell line showed that forskolin resulted in CREB phosphorylation at serine 133, typically associated with CREB activation, even when a PKA inhibitor was added. Therefore, EPAC was proposed as an upstream mediator for CREB activation rather than PKA in this cell line [15]. However, after forskolin addition, CREB activation in NS-1 cells was inhibited following PKA inhibition, while neurite outgrowth was still stimulated. These results suggest that in NS-1 cells, the effect on neurite outgrowth is both PKA- and CREB-independent. The forskolin-induced neuritogenesis is possibly EPAC-mediated, but this has not been investigated further. Moreover, cAMP analogs stimulated ERK phosphorylation in NS-1 cells independent of PKA, indicating that AC stimulation by forskolin and ERK are connected, leading to neurite outgrowth [112].

3.3. Summary

PDE4 inhibition has been demonstrated to stimulate structural neuroplasticity through EPAC-mediated CREB phosphorylation but not PKA, leading to CREB/BDNF/Arg1 pathway activation. Moreover, crosstalk with the Ras/ERK and PI3K/Akt pathways has been established through BNDF-mediated TrkB activation. PDE4 inhibition also blocks the Rho/ROCK pathway through cAMP/EPAC, thereby promoting axonal growth. There are contradictory results on PKA and EPAC dependency in the neurite outgrowth process, which can be explained by different methodologies, neuronal cell types and models, mechanisms of action of the used inhibitor/analog, animal models and administration routes, and thus the context. Moreover, research showed that EPAC activation is more sensitive to intracellular PDE4 levels, while PKA activation is more influenced by the present cAMP levels [113]. Consequently, these differences in sensitivity can potentially explain why PDE4 inhibition demonstrated promoted neuronal outgrowth through EPAC, whereas cAMP analogs proposed PKA-mediated neuronal elongation. However, evidence states that neurite outgrowth can be boosted independent of PKA upon PDE4 inhibition. cAMP elevations through cAMP stimulants or analogs have also been reported to enhance neuroplasticity through PKA and PI3K pathways. The role of the MAPK subfamilies upon cAMP increases is contradictory, although a role for ERK in neuroplasticity and synaptic transmission has been demonstrated[114]. Moreover, it must be kept in mind that cAMP elevations, either upon cAMP stimulation or PDE inhibition, might indirectly affect neuroplasticity and neurite outgrowth. Interference with processes such as cell proliferation, cell adhesion molecules, and extracellular matrix components could contribute to neuroplasticity (Fig. 3).

4. Neurotransmission and long-term potentiation

LTP can be defined as a strengthening of synaptic connections. Within the synaptic cleft, the space between two adjacent synapses, neurotransmitters are released to either activate or inhibit receptors, mediating synaptic connections. Dopamine, glutamate, and gamma-aminobutyric acid (GABA) are well-characterized neurotransmitters, the dysregulation of which is implicated as pivotal in various neurodegenerative diseases [115–122]. cAMP has been shown as a partial regulator of these three neurotransmission processes, with different PDEs implicated to cause distinct downstream effects [123–125]. Notably, LTP has been proposed as the primary mechanism behind memory and learning [126], and its disruption has been linked to AD [120]. cAMP and its PDE regulators have been shown to influence late-LTP in the hippocampus, contributing to changes in memory functions [127–129].

4.1. cAMP signaling upon PDE4 inhibition in neurotransmission

Considering dopamine transmission, PDE research has been primarily focused on PKA-mediated effects on the function of D1- and D2-type medium spiny neurons and subsequent phosphorylation of dopamine- and cyclic-AMP-regulated phosphoprotein (DARPP-32) [130]. *Mota et al.* showed that the PDE4 inhibitor rolipram upregulated DARPP-32 phosphorylation in D1 and D2 striatal spiny neurons in a PKA-mediated manner. The phosphorylation of DARPP-32 leads to a multitude of downstream effects, affecting, for example, GABAergic neurotransmission and ion channels, such as Na^+, through the inhibition of protein phosphatase-1 (PP-1) (the downstream targets of PP-1 are reviewed in depth in [131]. In the same study, PDE4 inhibition increased PKA-mediated phospho-Ser40 phosphorylation of presynaptic enzyme tyrosine hydroxylase (TH), suggesting that PDE4 could be involved in cAMP/PKA-mediated dopamine neurotransmission at presynaptic and postsynaptic terminals [132]. The D1 receptor activity was enhanced by rolipram-mediated PDE4 inhibition in frontal cortex pyramidal neurons, further confirming the potentiating role of cAMP/PKA-mediated DARPP-32 phosphorylation [133]. In this *in vivo* study, rolipram-treated animals had improved sensory-motor task execution, which was DARPP-32 dependent [133]. Together, these findings highlight the roles of PDE4 in regulating dopamine transmission via the modulation of the cAMP/PKA signaling cascade and the subsequent regulation of DARPP-32 and TH phosphorylation. Nevertheless, the downstream effects of DARPP-32 phosphorylation have not yet been explored. DARPP-32 has been shown to indirectly inhibit CRE, which results in CREB dephosphorylation and subsequent transcriptional changes [134]

PKA appears to be the most important downstream target of PDE4 inhibition on glutamate neurotransmission. PDE4 inhibition by rolipram was positively correlated with increased glutamate release through the enhanced activation of the cAMP/PKA pathway and subsequent potentiation of 4-aminopyridine (4AP), which caused a dose-dependent increase in Ca^{2+}-mediated glutamate release [135]. The role of PKA in the process was deducted by either inhibiting PKA, which interfered with glutamate release, or directly potentiating it by PKA activation, which exacerbated rolipram's effects. Interestingly, an *in silico* study demonstrated the critical role of ERK-mediated cAMP/PKA regulation and

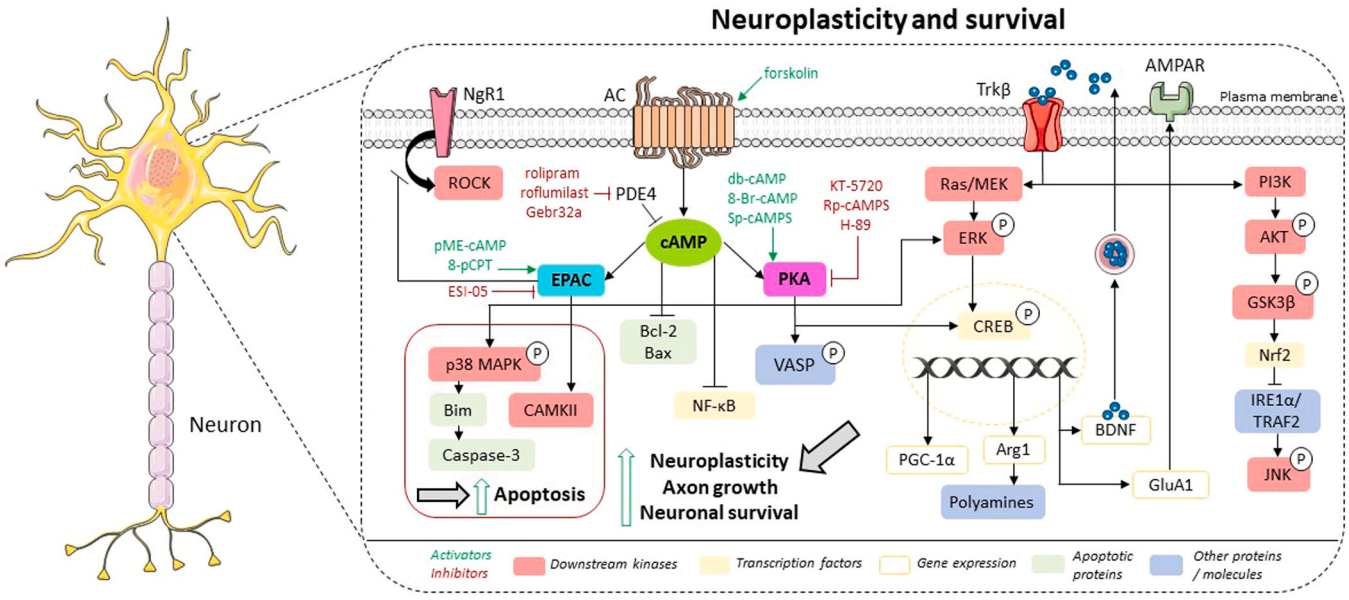

Fig. 3. : Downstream signaling upon cAMP stimulation or PDE4 inhibition in neuronal plasticity and survival**.** Promoting cAMP signaling through the inhibition of PDE4 (rolipram, roflumilast, Gebr32a) or cAMP analogs specific for PKA (db-cAMP, 8-Br-cAMP, or Sp-cAMPS) and/or EPAC (pME-cAMP or 8-pCPT) exerts different effects in neuronal plasticity and survival. PDE4 inhibition stimulates neuroplasticity through EPAC-, but not PKA-, mediated CREB phosphorylation, leading to the gene expression of BDNF and Arg1. Additionally, EPAC can promote neuronal plasticity by inhibiting NgR1-induced ROCK signaling. BDNF can activate Trkβ transduction to induce Ras/MEK signaling and promote neuroplasticity. Ras/MEK transduction can crosstalk with PI3K/AKT/GSK3β/Nrf2/IRE1α/TRAF2/JNK signaling, which promotes neuronal survival. However, cAMP analogs specific for PKA suggest PKA as an important downstream modulator for neuronal plasticity. PKA mediates its effect through phosphorylation of CREB or indirectly by ERK phosphorylation, increased PGC-1α, Arg1, BDNF, and GluA1 expression. GluA1 receptor expression can, in turn, promote AMPAR expression to stimulate neuronal signaling. Moreover, PKA-mediated inhibition of apoptotic proteins NF-κB, Bcl-2, and Bax attenuate neuronal apoptosis. In contrast, cAMP analogs for EPAC promote apoptosis by p38 MAPK/Bim/Caspase-3 and EPAC-mediated CAMKII signaling. Therefore, EPAC is suggested as the main modulator of cAMP-mediated apoptosis. *NgR1, Nogo receptor 1; AC, Adeylyl cyclase; TrkB, Tyrosine kinase B; AMPAR, α-Amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid receptor; AC, Adeylyl cyclase; PDE4, Phosphodiesterase 4; cAMP, Cyclic adenosine monophosphate; PKA, Protein kinase A; db-cAMP, Di-butyryl-cyclic AMP; 8-Br-cAMP, 8-Bromo-cyclic AMP; Sp-cAMPS; Sp-adenosine 3′,5′-cyclic monophosphorothioate triethylammonium salt hydrate; Rp-cAMPS, Rp-adenosine 3′,5′-cyclic monophosphorothioate triethylammonium salt; Bcl-2, B-cell leukemia/lymphoma 2 protein; Bax, Bcl-2-associated X protein; NF-κB, Nuclear factor κ B; VASP, Vasodilator-stimulated phosphoprotein; CREB, cAMP-response element binding protein; MEK, Mitogen-activated protein kinase; ERK, Extracellular signal-regulated kinase; PI3K, Phosphoinositide 3-kinase; AKT, Protein kinase B; GSK3β, Glycogen synthase kinase-3 β; Nrf2, Nuclear factor erythroid 2; IRE1α, inositol-requiring enzyme 1 α; TRAF2, TNF receptor-associated factor 2; JNK, c-Jun N-terminal kinase; PGC-1α, Peroxisome proliferator-activated receptor-gamma coactivator; Arg1, Arginase 1; BDNF, Brain derived neurothrophic factor; GluA1, Glutamate receptor 1; EPAC, Exchange protein directly activated by cAMP; pME-cAMP, 8-pCPT, 8-(4-chlorophenylthio)-2′-O-methyl-cyclic AMP; 8-(4-chlorophenylthio)-2′-O-methyl-cyclic AMP; p38 MAPK, Mitogen-activated protein kinase; Bim, Bcl-2-interacting mediator; ROCK, Rho-associated coiled-coil kinase; CAMKII, Calmodulin-dependent protein kinase II.*

subsequent GluA1 phosphorylation. Here, PDE4 was shown to be a critical mediator between PKA and ERK since ERK phosphorylation inhibits some of the PDE4 isoforms [136]. According to the conducted literature search, no research concerning PDE4 inhibition's effect on GABAergic neurotransmission exists.

In the realm of late-LTP, research predominantly centers around the PDE4 family. Research showed that repeated stress lowered the LTP threshold, leading to decreased PDE4 expression in amygdala neurons. When the same neurons without stress exposure were treated with rolipram, the effects of the stress were mimicked, suggesting a pivotal role for PDE4 in regulating LTP induction sensitivity via the cAMP/PKA pathway [137]. Furthermore, studies in hippocampal CA1 slices have shown a temporal variation of the PDE4B3 isoform expression during LTP activation [138]. Other studies employing PDE4 inhibitors seem to confirm the PDE4 role in activating LTP via cAMP regulation. For instance, *in vivo* studies have revealed that rolipram treatment in combination with forskolin in murineCA1 slices results in LTP potentiation [139]. Another study on CA1 slices has found that brain slices of *Pde4d* knockout (KO) mice displayed elevated LTP compared to the control counterparts [140]. Treatment with rolipram has also been shown to reverse object memory deterioration in rats previously treated with an NMDA blocker and promoted the transition from short-term potentiation (STP) into LTP in healthy animals [141]. However, most of these studies implicate that LTP activation is regulated via the cAMP/PKA pathway by simply referring to the previous literature. Confirming these findings through PKA inhibition was only performed by *Ryan et al.* [137]. Recently, *in silico* findings indicated a role for ERK in the temporal regulation of LTP, pointing to the need for further evaluation of LTP-inducing mechanisms [142]. Finally, it has been shown that Aβ-induced inhibition of LTP in the hippocampus may be reversed by applying rolipram *ex vivo*, which is particularly interesting in AD. More specifically, it was shown that the PDE4 inhibition in brain slices exposed to soluble oligomers Aβ prevented the Aβ-induced inhibition of LTP. PKA was suggested to be involved, as its inhibition blocked LTP in the CA3 mossy fibers [143]. Unfortunately, most studies above did not explore the downstream cascade activated upon PDE4 inhibition beyond PKA.

4.2. cAMP signaling upon cAMP stimulants or cAMP analogs in neurotransmission

cAMP has been investigated in many different aspects and stages of dopamine neurotransmission. In D1 rat neurons, suppressing the cAMP/PKA pathway through a cAMP antagonist reversed the inhibition of Ca^{2+}-dependent K^{+} currents and subsequently increased spiking frequency. Interestingly, none of the main cAMP effectors (PKA/EPAC) seemed responsible for K^{+} current regulation [16]. In the D2 neurons, cAMP antagonism *in vivo* has been shown to inhibit the H3 phospho-acetylation, and conversely, stimulation of cAMP/PKA activity has enhanced the process. These findings suggest a role for cAMP and

PKA in regulating chromatin modifications in the D2 neurons and subsequent changes in dopamine neurotransmission [144]. Treatment with 8-Br-cAMP or forskolin has been shown to partially inhibit the activity of rat D2 neurons, which was reversed upon PKA inhibition, suggesting the role of PKA in the process. [145]. Additionally, rat striatal synaptosomes with 8-Br-cAMP significantly increased the dopamine transporter uptake and, subsequently, increased dopamine transmission in a PKA-dependent manner. Yet downstream molecules of PKA that promote dopamine transporter activity have not been studied [146]. In future research, it will be essential to investigate whether similar effects on dopamine transmission can be obtained via PDE inhibition rather than direct cAMP stimulation/inhibition and look further into which mechanisms are responsible for those effects.

In terms of glutamate neurotransmission, a study in the early 90 s has provided insight into cAMP stimulation and non-NMDA-type channel activity. It was revealed that treating hippocampal pyramidal neurons with forskolin increased the average opening time of these channels through a PKA-dependent process [123]. A few years later, a similar relation was found in synaptosomes obtained from cerebrocortical neurons, where forskolin treatment increased the facilitation of glutamate release. This was also hypothesized to be cAMP/PKA-mediated as PKA inhibition reversed the effects. Similar to the PDE inhibition study by Wang [135], PKA potentiates the effects of 4AP, causes channel opening, and increases the amount of glutamate released. However, the interaction between PKA and 4AP was not explored further [135,147]. More recently, it has been suggested that EPAC is also involved in increasing glutamate release. Using the EPAC agonist 8-CPT-cAMP while blocking PKA increased glutamate release. Moreover, EPAC was found to trigger glutamate release through PLC activation, leading to Munc13–1 translocation, which is critical for neurotransmission, and increased Rab3A/RIM1α association [148]. Clearly, there is evidence that cAMP mediates neuronal excitability. In astrocytes, treatment with db-cAMP has been shown to upregulate L-[3 H]glutamate through increased Na^+ channel stimulation [149,150]. In particular, the cAMP stimulation caused an increase in glutamate transporter-1 (GLT-1) protein expression and glutamate aspartate transporter (GLAST) abundance, both responsible for glutamate transport in the glial cells. Here, PKA was not correlated to transporter upregulation, but no other downstream targets were explored [150]. The results of these studies are supported by another study that used 8-Br-cAMP and showed increased glutamate synthesis in glial cells [151].

The effect of cAMP on GABAergic neurotransmission is the least studied out of the three neurotransmitters discussed [152–154]. Forskolin treatment increased phosphorylated β1-adrenergic receptors through PKA and led to a weaker response of the receptors to GABA stimulation, indicating increased desensitization of rat-derived neuronal cells [153]. Additionally, forskolin was found to suppress postsynaptic GABA receptors in fruit flies, decreasing the synaptic excitability of the treated neurons. This process was determined to be PKA-dependent, as the inhibition of PKA reversed the inhibition of GABAergic currents. [155]. Recently, it has been suggested that even though PKA is the main effector of $GABA_A$ receptor phosphorylation, EPAC seems to be the main reason for the phosphorylation of this receptor at the mixed inhibitory synapses. Forskolin-induced phosphorylation of gephyrin, a scaffold protein present at synapses and a common binding site for glycerin and $GABA_A$ receptors, was found to be regulated by EPAC in primary neuronal rat cells. Additionally, EPAC stimulation increased the mobility of $GABA_A$ receptors at the synapses by increasing the binding strength between $GABA_A$ and gephyrin [156].

Research on the impact of cAMP on LTP has focused on understanding the effect of cAMP stimulants on CA1 neurons, which are crucial for memory formation [157]. It was revealed that cAMP stimulation can mimic the pattern of late LTP, while cAMP antagonism can block it upon its induction in the hippocampal slices. The LTP induction was considered PKA-dependent, yet further exploration of downstream processes responsible for LTP induction is necessary [158,159]. These *in vitro* studies were translated into *in vivo* studies in mice, where intracranial forskolin application to the CA1 area restored long-term memory (LTM) performance [160]. While initial studies in the area of cAMP-influence on LTP suggested that PKA is responsible for late LTP induction, later studies suggested the involvement of ERK and CREB in the process, specifically in CA1 neurons. More precisely, the application of forskolin and simultaneous blocking of MEK hindered LTP induction in these brain slices but not in the CA3 area [161]. It was also shown that without NMDA channel activation, cAMP-induced LTP was not established in the CA1 region [162]. More recently, it was demonstrated that cAMP has different roles in LTP induction in other cell types. Inhibition of cAMP activity was indeed found to alter the LTP pattern in bursting, which was PKA mediation, but not in regular firing cells [163]. In the context of AD, cAMP potentiators have been found to reverse LTP inhibition. It is suggested that Aβ-induced-induced LTP inhibition is caused by β1-adrenergic receptor activation in hippocampal microglia cells, whereby forskolin can reverse this effect through PKA [143,164,165]. The only downstream target analyzed in this case was CaMKII, whose inhibition did not reverse the LTP induction potentiated by PKA. This means that there must be another pathway explaining PKA's effect on LTP [164]. As briefly mentioned earlier in the PDE inhibition context, looking at the CA3 mossy fibers specifically revealed that both the β1- and β2-adrenergic receptors may be involved in the LTP formation, and their inhibitory effect on LTP can be reversed by the application of forskolin [143]. In both cases, PKA is suggested as a downstream effector of LTP induction [143,164]. Interestingly, EPAC was also shown to be responsible for LTP induction in CA1 neurons. The application of EPAC-specific cAMP analog 8-CPT-cAMP and the concurrent application of a PKA inhibitor to hippocampal mouse slices resulted in LTP induction. The effect was ERK-mediated, as MEK inhibition reversed the LTP maintenance obtained by applying 8-CPT-cAMP. Furthermore, it was suggested that EPAC forms the link between cAMP and ERK pathways that induce LTP [166].

4.3. Summary

Considering neurotransmission, PDE4 inhibition has been demonstrated to increase dopamine release through cAMP/PKA-mediated phosphorylation of DARPP-32 and TH and subsequent inhibition of PP-1. Conversely, cAMP elevations upon cAMP analog treatment have been shown to attenuate dopamine transmission by modulating dopamine transporter capacity through a PKA-dependent process. Moreover, PDE4 inhibitors have been found to stimulate glutamate release through cAMP/PKA, potentiating the effects of 4AP. Similarly, cAMP stimulators increased non-NMDA channel opening and glutamate release through the same pathway. Additionally, EPAC and PLC have been found to stimulate glutamate release. cAMP elevations also promote glutamate transport in glial cells through a PKA-independent process. To our knowledge, the effect of PDE4 inhibition on GABAergic transmission has yet to be studied, rendering a knowledge gap. Elevations in cAMP levels through cAMP analogs indicate a role for both PKA and EPAC in GABAergic transmission. Lastly, PDE4 inhibition has been shown to promote LTP, and thus memory and learning processes, through the cAMP/PKA pathway. cAMP/ERK has also been indicated in this process, but exact downstream mechanisms remain elusive (Fig. 4).

5. Neuronal survival

Neuronal survival is mediated by a complex interplay between glial cells and the CNS environment. Neurodegenerative disorders are characterized by changing CNS conditions leading to neuronal degeneration, synaptic loss, and, ultimately, neuronal death [167]. Promoting neuronal survival has already been shown to improve disease outcomes in AD, PD, ALS, and traumatic CNS injuries [168–171]. Multiple cAMP analogs, as well as PDE4 inhibitors, are demonstrated to promote neuronal survival and are therefore considered promising therapeutic

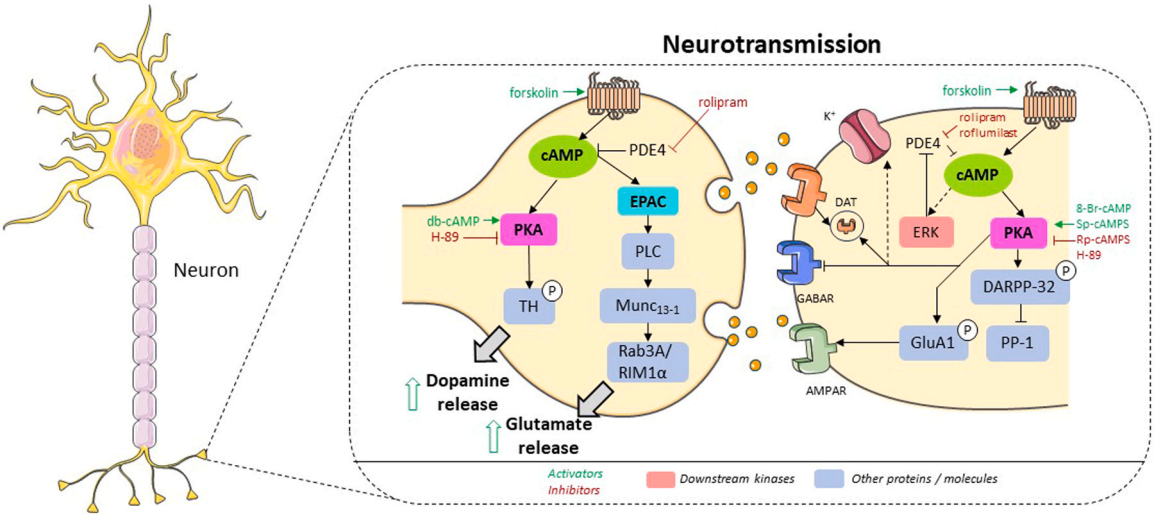

Fig. 4. : Downstream signaling upon cAMP stimulation or PDE4 inhibition in neurotransmission**.** Promoting cAMP signaling by inhibiting PDE4 (rolipram or roflumilast) or cAMP analogs specific for PKA (db-cAMP, 8-Br-cAMP or Sp-cAMPS) and/or EPAC stimulates neurotransmission. PDE4 inhibition promotes dopamine release through PKA/TH. In contrast, cAMP analogs for PKA reduce dopamine signaling by promoting dopamine receptor transport from the plasma membrane to the intracellular environment. PDE4 inhibitors promote glutamate release via EPAC/PLC/Munc13–1/Rab3A/Rim1α signaling. cAMP analogs for PKA stimulate the expression of neurotransmitter receptors GABA and AMPA and modulate receptor gating of neurotransmitter receptors through inhibition of PP-1 via DARPP-32 and potentially also of potassium channels. *AC, Adeylyl cyclase; PDE4, Phosphodiesterase 4; cAMP, Cyclic adenosine monophosphate; PKA, Protein kinase A; db-cAMP, Di-butyryl-cyclic AMP; 8-Br-cAMP, 8-Bromo-cyclic AMP; Sp-cAMPS; Sp-adenosine 3c′,5c′-cyclic mono phosphorothioate triethylammonium salt hydrate; PKA, Protein kinase A; TH, Tyrosine hydroxylase; EPAC, Exchange protein directly activated by cAMP; PLC, Phospholipase C; Rab3A, Ras-related protein Rab-3A; RIM, Rab3-interacting molecule; ERK, Extracellular signal-regulated kinase; DARPP-32, dopamine- and cyclic-AMP-regulated phosphoprotein of molecular weight 32,000; PP-1, Phosphatase-1; GluA1, Glutamate receptor 1; DAT, Dopamine transporter; GABAR, Gamma-aminobutyric acid receptor; AMPAR, α-Amino-3-hydroxy-5-methyl-4-isoxazole-propionic acid receptor.*

strategies for neurodegenerative disorders.

5.1. cAMP signaling upon PDE4 inhibition in neuronal survival

PDE4 inhibition is widely recognized for promoting both *in vitro* and *in vivo* neuronal survival. In a 3xTg AD mouse model, administration of the PDE4 inhibitor rolipram increased the expression of the anti-apoptotic protein Bcl-2 and decreased the expression of the pro-apoptotic protein Bax in the prefrontal cortex and hippocampus. This was accompanied by an increase in the number of neurons in the hippocampus. PKA and EPAC were both upregulated upon rolipram treatment, as well as downstream proteasome molecule 26 S and ERK, respectively [172]. The upregulation of PKA/26 S and EPAC/ERK suggests the involvement of both signaling cascades in regulating neuronal survival. Indeed, rolipram reduced neuronal death in the human neuroblastoma SH-SY5Y cell line, while its neuroprotective effect was reversed upon PKA inhibition [173].

GSK3β is postulated as an important downstream molecule of PKA to execute the neuroprotective effects upon PDE4 inhibition. PKA can phosphorylate GSK3β directly or modulate GSK3β activity via PI3K [174,175]. PDE4 inhibition with roflumilast prevented cell death in the HT-22 neuronal cell line, reversed decreased AKT expression, and promoted GSK3β phosphorylation. However, GSK3β inactivation by inducing a mutation in serine 9 (S9A) abolished these effects, which implies that this region in GSK3β is demanded for the beneficial actions of roflumilast on neuronal survival [176]. Importantly, AKT is an established downstream signaling molecule of EPAC and PI3K and an upstream regulator of GSK3β [174,175,177]. Phosphorylated GSK3β can activate inositol-requiring transmembrane kinase/endoribonuclease 1α (IRE1α), which in turn interacts with TNF-receptor-associated factor 2 (TRAF2), leading to the activation of the JNK pathway to induce apoptosis [178]. The inhibition of PDE4 with roflumilast disrupted IRE1α and TRAF2 colocalization and promoted antioxidant regulator Nrf2. The disruption of this interaction is hypothesized to be the underlying mechanism that reduces JNK phosphorylation upon PDE4 inhibition. Consequently, PDE4 inhibition was postulated to promote neuronal survival by enhanced GSK3β phosphorylation, leading to Nrf2-mediated reductions in oxidative stress and, hence, disrupted IRE1α/TRAF2 interaction, resulting in ceased JNK signaling [176]. Remarkably, this is one of the few studies that thoroughly investigated the downstream mechanism of PDE4 inhibition. The involvement of JNK signaling in neuronal survival was further supported by *Xiao et al.*, who demonstrated that PDE4 inhibition in HT-22 cells diminished TNF-α-induced neuronal cell death through decreased JNK phosphorylation [179]. Moreover, the translocation of p65, a dimer of NF-κB signaling, was attenuated upon PDE4 inhibition, suggesting a potential role of NF-κB in neuronal cell death. Thus, inhibition of the JNK and NF-κB pathways following PDE4 inhibition promotes neuronal survival [179].

Furthermore, PDE4 inhibition with roflupram and rolipram was found to exert neuroprotective effects *in vitro* and *in vivo* via PKA/CREB signaling [180]. Pretreatment with roflupram opposed phenylpyridinium iodide (MPP+)-induced apoptosis and promoted CREB phosphorylation, whereby these effects were blocked upon PKA inhibition in SH-SY5Y cells. The neuroprotective effect of CREB can potentially be mediated by increasing the gene expression of PGC-1α, which is involved in antioxidant mechanisms [181]. Indeed, PGC-1α activity was increased upon PDE4 inhibition, and blocking PGC-1α opposed the beneficial effects of rolipram. Additionally, roflupram or rolipram promoted neuronal survival and increased pCREB levels in the striatum and substantia nigra in mice pretreated with a neurotoxic compound [180]. The role of CREB in stimulating neuronal proliferation was further explored in an ischemic mouse model. Rolipram treatment increased neuronal proliferation and CREB phosphorylation, but when pCREB signaling was inhibited, the number of newborn neurons decreased. Consequently, a significant role of CREB signaling in promoting neuronal survival is suggested [182]. Notably, low doses of rolipram promoted neuronal survival, while higher doses did not. BDNF administration alone also promoted neuronal viability, while BDNF and a low dose of rolipram could further enhance neuronal survival. Moreover, rolipram treatment alone promoted ERK and CREB phosphorylation [183]. Therefore, it was postulated that PDE4 inhibition stimulates

neuronal survival through ERK/CREB signaling.

5.2. cAMP signaling upon cAMP stimulants or cAMP analogs in neuronal survival

The downstream signaling cascade of cAMP/PKA and cAMP/EPAC and their influence on neuronal survival can be further elucidated using cAMP analogs. Stimulation of cAMP signaling with forskolin in primary rat interneurons could oppose neuronal apoptosis. PKA/GSK3β was proposed as the underlying mechanism since cAMP stimulation decreased GSK3β activity, a response that was neutralized upon PKA inhibition. Moreover, MEK inhibition did not affect GSK3β signaling nor alter neuronal apoptosis [184]. Therefore, it was hypothesized that the neuroprotective effect of cAMP was mediated through PKA/GSK3β signaling but not through the MEK/ERK pathway. Additionally, the PKA-specific cAMP analog Sp-cAMPS reduced neuronal cell death by upregulating pCREB in rats. Consequently, PKA can potentially mediate its neuroprotective effects through CREB signaling [185]. In contrast, high concentrations of forskolin reduced neuronal survival in primary rat motor neurons [186]. BDNF is known to stimulate neuronal survival. However, the neuroprotective effect of BDNF in apoptotic conditions in rat-derived neuronal cells was nullified upon 10 μM forskolin. Forskolin-induced apoptosis at high concentrations is likely mediated through PKA since PKA inhibition could prevent apoptosis during BDNF stimulation. Forskolin administration decreased PI3K-mediated AKT phosphorylation and stimulated MAPK phosphorylation. These findings imply that cAMP-mediated apoptosis can be induced through PKA/-MAPK signaling [187]. Nevertheless, it should be noted that a study by *Kranz et al.* concerning PDE4 signaling and apoptosis revealed that low rolipram doses promote cellular survival together with BDNF [183]. In contrast, high doses of rolipram (1–10 μM) have the opposite effect. Therefore, the results of cAMP stimulation on neuronal apoptosis through PKA signaling may depend on the concentration of cAMP and the cell type with its signaling molecules.

Past research has proven that cAMP can also mediate neuronal apoptosis through EPAC signaling since EPAC stimulation increases neuronal apoptosis in mouse cortical cells. EPAC agonism also increased p38 MAPK phosphorylation and the levels of the apoptotic stimulating molecule Bcl-2-like protein 11 (Bim) but not JNK signaling. Inhibition of p38 MAPK signaling could abolish EPAC-mediated increases in Bim expression. Lastly, EPAC stimulation promoted Bim-Bcl2 interaction, and Bim silencing could oppose EPAC-mediated neuronal apoptosis. Hence, EPAC is suggested to mediate neuronal apoptosis through the EPAC/p38 MAPK/Bim/Bcl2 signaling cascade [188]. These results were supported *in vivo* using a rat model of intracerebral hemorrhage. Here, EPAC2 inhibition decreased neuronal apoptosis. In addition, p38, Bim, and caspase-3 expression levels were reduced in neurons upon EPAC2 inhibition, suggesting p38 MAPK/Bim/caspase-3 as the underlying pathway for EPAC-induced apoptosis [189]. The EPAC/p38 MAPK pathway was further suggested *in vivo* in the context of TBI. After TBI, Epac2 expression was increased in mice. Additionally, pretreatment with an EPAC2 antagonist decreased neuronal death and reduced p38 phosphorylation [190]. In contrast, after retinal ischemia-reperfusion injury, not Epac2 but Epac1 and its downstream molecules Rap1-Gtp, Akt, and Erk were increased. Interestingly, the EPAC-stimulating cAMP analog 8-CPT-cAMP induced neuronal cell death in retinal ganglion cells but not in Epac1 KO cells, indicating that predominantly Epac2 signaling is responsible for neuronal apoptosis [191]. Research suggests that, indeed, EPAC2 but not EPAC1 is colocalized with PKA and AKT in cortical neurons [192]. Application of a CaMKII inhibitor opposed neuronal cell death upon 8-CPT-cAMP administration, while inhibitors for p38, MAPK, JNK, PKC, or PI3K/AKT could not prevent EPAC-mediated cellular apoptosis. Consequently, CaMKII has been postulated as another important downstream molecule in EPAC signaling that promotes cellular death. Notably, this research attributed the pro-apoptotic properties of EPAC signaling to EPAC1 rather than EPAC2, as stated above [191].

5.3. Summary

In conclusion, neuronal survival is predominantly mediated through PKA signaling with downstream molecules GSK3β/Nrf2 and CREB/PGC-1α. Although PDE4 inhibition was thought to promote neuronal survival via EPAC, accumulating evidence states that EPAC signaling by cAMP elevations promotes apoptosis via CaMK and p38 MAPK/Bim/Bcl-2/caspase 3. These contradictory effects can be attributed to varying cAMP levels based on cell type, developmental stage, concentration and duration of cAMP treatment, intensity of cAMP elevation, and signaling pathway crosstalk. Consequently, the PDE4 inhibitor dose plays an essential role in the observed outcome, and more research is necessary to confirm the beneficial effects of increased cAMP levels on neuronal survival (Fig. 3).

6. Myelination

Another process crucial for proper CNS functioning in which cAMP plays a pivotal role is oligodendrocyte development and myelination. The primary function of oligodendrocytes, the myelinating cells of the CNS, is to produce a lipid-rich substance called myelin, which electrically insulates neuronal axons. The myelin sheath is critical for safeguarding neurons and facilitating rapid and efficient signal transmission [8,193]. In demyelinating disorders such as MS and other neurodegenerative conditions marked by oligodendrocyte loss, failure of OPC differentiation leads to chronic demyelination and axonal damage, culminating in neurodegeneration. Since anti-inflammatory drugs only target one aspect of demyelinating diseases and fail to reverse disease progression, there is an urgent need for myelin-regenerative therapies. Considering that cAMP plays a crucial role in OPC differentiation, elevating its levels through PDE inhibition is a promising therapeutic approach that we, and others, have been investigating over the last decade [8,26,194].

6.1. cAMP signaling upon PDE4 inhibition in myelination

PDE4 inhibitors have been shown to enhance OPC differentiation to mature myelinating oligodendrocytes and improve remyelination in the context of MS, as demonstrated by us and others, but also in other disease contexts like SCI [26,84,194–196]. However, downstream signaling pathways activated in OPCs and oligodendrocytes upon cAMP elevations induced by PD4E inhibition have not yet been adequately investigated.

The PDE4 inhibitor rolipram promotes oligodendrocyte differentiation and maturation of primary mouse OPCs. It also increased remyelination in a cuprizone-induced demyelination mouse model. Rolipram treatment increased ERK phosphorylation, whereas MAPK inhibition partially blocked its effects, indicating that rolipram acts partly through the MAPK/ERK pathway. However, it was not investigated whether these effects were PKA-dependent [195]. Transcriptional profiling on primary rat OPCs also identified MAPK signaling as an important mediator of OPC differentiation upon PDE4 inhibition. More specifically, rolipram significantly increased the phosphorylation of ERK1/2, p38 MAPK, and their downstream signaling molecule CREB. Hence, a signaling pathway for CNS remyelination was proposed where elevated levels of cAMP cause ERK1/2 and p38 MAPK phosphorylation through direct interaction or indirect activation of PKA, triggering the activation of CREB and subsequent OPC differentiation. Nevertheless, PKA levels or expression of genes regulated by CREB were not investigated[194].

In an *in vitro* SCI model, researchers found that inhibition of the PDE4 HARBS conformer using R-rolipram promotes myelination through EPAC activation instead of PKA. On the contrary, high concentrations of rolipram did not enhance myelination because of an inhibitory action mediated by PKA, demonstrating a detrimental role of PKA in

myelination. Hence, rolipram-induced myelination may result from a pool of cAMP controlled by a PDE4 subpopulation that adopts the HARBS conformation and can boost myelination through EPAC [84]. This supports the notion that spatially different pools of cAMP are regulated by distinct forms of PDE(4)s and cause multiple physiological effects through separate downstream effectors, i.e., PKA or EPAC [84,197,198]. Yet, the involvement of EPAC upon PDE4 inhibition has not been thoroughly studied. Further investigation into specific genes of the PDE4 family (i.e., PDE4A-D) revealed that selective PDE4D inhibition using Gebr32a promotes OPC differentiation and remyelination. Moreover, it was conceptualized that short PDE4D isoforms (i.e., PDE4D1/2 and PDE4D6) are key targets to induce myelination. However, the signaling pathway activated upon Gebr32a-mediated PDE4D inhibition remains to be explored [26].

6.2. cAMP signaling upon cAMP stimulants or cAMP analogs in myelination

Studies in the early 1980s already showed that oligodendrocyte differentiation can be induced by elevating the intracellular concentration of cAMP in primary rat brain cells. By examining the expression of oligodendrocyte markers and myelin components like MBP, 2',3' cyclic nucleotide 3'-phosphohydrolase (CNP), it was found that cAMP analogs accelerate the expression of these components and thus oligodendrocyte differentiation. Since 8-Br-cAMP specifically leads to PKA activation, a potent role of PKA in oligodendrocyte differentiation could be suggested [199,200]. A decade later, cAMP was confirmed to regulate OPC proliferation and differentiation. Upon cAMP elevations induced by db-cAMP, OPC proliferation and differentiation were inhibited and stimulated, respectively [17]. Other studies suggest that cAMP-dependent induction of oligodendrocyte differentiation is at least partly mediated by CREB [201,202]. Treating rat oligodendrocytes with db-cAMP and CREB antisense oligomers led to less differentiation than db-cAMP alone, and expression of myelin proteolipid protein (Plp) was increased despite inhibition of Creb expression [201,202]. Hence, this data suggests that CREB plays a role in the cAMP-dependent upregulation of Mbp, whereas it does not mediate the cAMP-dependent stimulatory effects on Plp expression. These findings indicate that multiple downstream signaling molecules mediate the stimulation of oligodendrocyte differentiation and myelination by cAMP. Moreover, CREB was believed to regulate Mbp gene expression indirectly since there is a long lag time required for cAMP-dependent stimulation of Mbp expression, and it was thought that the Mbp gene region does not have a CREB-binding site [202]. These studies did not investigate the kinase responsible for phosphorylating and activating CREB, which is primarily carried out by PKA but also by CaMK, PKC, or members of the MAPK family [29,32,202]. Interestingly, a later study in HEK293T cells showed that the *MBP* promotor region does have a CREB-binding site [203]. Another study demonstrated that cAMP/PKA pathway stimulation through forskolin or db-cAMP induces the expression of Cnp and Plp in Oli-neu cells and primary rat OPCs. However, the cAMP agonists exhibited no significant effect on MBP production and only partially induced phenotypic differentiation. Additionally, a pathway analysis that pointed to the involvement of ERK, hypothesized through cAMP crosstalk via EPAC. This indicates that PKA is only one of many partially redundant pathways leading to differentiation [204]. *Sato-Bigbee et al.* suggest that CREB plays different regulatory roles at distinct oligodendrocyte development stages, coupled with specific signaling pathways and kinases [205]. CREB phosphorylation can be mediated by both elevated levels of cAMP and Ca^{2+}, mediated by PKA and PKC, respectively, in OPCs and young oligodendrocytes. In the early stages of immature OPCs, CREB is proposed to regulate cell proliferation, and its phosphorylation is mediated by PKC and MAPK pathways. In contrast, when cells are already committed oligodendrocytes, CREB phosphorylation is mediated by PKA and involved in Mbp gene expression. These conclusions were drawn upon treating oligodendrocytes with db-cAMP and various kinase inhibitors for PKC, PKA, CaMK, and MEK [202,205]. Other studies confirm a similar idea but with opposing signaling pathways. They demonstrate that CREB phosphorylation, primarily mediated by PKA, advances the differentiation of OPCs to immature oligodendrocytes [29]. In contrast, PKC-mediated CREB phosphorylation boosts the differentiation from immature into mature oligodendrocytes [206]. Hence, oligodendrocytes might apply different CREB-phosphorylation systems (i.e., a PKA and PKC cascade) at distinct oligodendrocyte differentiation stages [29,206].

The complexity of cAMP signaling in the CNS becomes even more apparent when crosstalk with other pathways, like the MAPK pathways, is considered. *Bhat et al.* showed that by treating OPCs with a p38 MAPK inhibitor, forskolin-induced CREB phosphorylation was blocked, and forskolin-mediated stimulation of MBP expression was suppressed [32]. This suggests potential crosstalk between the cAMP and p38 MAPK pathway in the induction of myelin gene expression in differentiating oligodendrocytes. In addition, the authors suggest that CREB might be phosphorylated by downstream kinase targets of p38 MAPK, i.e., MAPKAP K-2, MNK1, or MSK-1, instead of by p38 MAPK directly. Yet, this was not investigated [32]. In a study using shi/shi OPCs from shiverer mutant mice carrying a recessive mutation in the MBP gene that disrupts myelination, OPC differentiation was accelerated by db-cAMP treatment. cAMP agonism activated JNK but did not alter the phosphorylation of p38 MAPK. Additionally, phosphorylation of transcription factor ATF-2 was observed, which may lead to increased expression of myelin-associated glycoprotein (Mag) [207]. In summary, multiple studies point to the involvement of the MAPK family in oligodendrocyte differentiation, possibly as a downstream signaling molecule activated upon cAMP elevations. The cAMP/MAPK crosstalk is better defined in neurons, where it has been demonstrated that ERK inhibits PDE4, thus preventing cAMP degradation and causing an ERK-dependent activation of the cAMP-PKA pathway [208]. In return, elevated cAMP can also activate ERK through EPAC-mediated activation of the Rap-1/B-Raf pathway, leading to ERK phosphorylation, highlighting the complex crosstalk between the two signaling pathways [209].

Other studies have indicated a role for both PKA and EPAC in oligodendrocyte maturation, differentiation, and myelination. Stimulation of Gαi/o-coupled GPCR17, mainly expressed in OPCs and involved in oligodendroglial maturation inhibition, decreased the activity of the AC-cAMP-PKA-CREB pathway and decreased EPAC activation in primary rat oligodendrocytes. The application of cAMP analog 8-CPT-cAMP restored oligodendrocyte maturation, indicating a role for cAMP/PKA and cAMP/EPAC in oligodendrocyte maturation. Thus, PKA and EPAC are potential therapeutic targets to boost remyelination in demyelinating diseases like MS since GPCR17 is abundantly expressed in white matter plaques of MS patients [193]. Another Gαi/o-coupled GPCR, the GPR37, predominantly expressed in mature oligodendrocytes, is a negative regulator of late-stage differentiation and myelination. This inhibitory action is mediated *in vitro* and *in vivo* by suppressing cAMP/EPAC/Raf signaling and subsequent loss of downstream ERK1/2 phosphorylation. In addition, GPCR37 attenuates the expression of the stimulatory myelin regulatory factor, which stimulates the expression of myelin genes. Interestingly, it was found that non-selective PDE inhibitor IBMX enhanced the OPC differentiation, similar to the levels compared to that observed in GPR37 KO cells [210]. Yet, a recent *in vivo* study employing Epac1/2 double-KO mice demonstrated more myelin sheaths, increased expression of myelin-related proteins, and increased OPC proliferation, indicating EPAC as a negative regulator of CNS myelination. The effect of EPAC1/2 on myelin development was thought to be age-dependent since its deficiency resulted in more myelin sheaths in the cerebral cortex in early but not late adult mice. Similarly, Epac KO enhanced the expression of Mag, Cnp, and Mbp in the cerebral cortex of young mice but not in old mice. These results indicate that EPAC could delay myelination rather than prevent it [211].

Notably, a study by *Malone et al.* [212] found that stimulation of cAMP in neurons can also enhance myelination. It has been shown

before that neuronal activity, specifically action potential propagation, mimicked by electrical stimulation (ESTIM), can promote OPC differentiation and myelination [213]. ESTIM applied to DRG neurons enhanced neuronal cAMP levels. Treatment of co-cultures of DRG and rat OPCs and pre-treatment of DRGs alone with forskolin, rolipram, and db-cAMP resulted in enhanced myelin segment formation in the absence of ESTIM. Moreover, inhibition of cAMP and PKA during ESTIM application hindered ESTIM-promoting effects on myelination. It is suggested that neuron-derived intracellular cAMP signaling regulates activity-dependent axon myelination by promoting axon receptivity to myelination [212]. However, they did not explore downstream cAMP signaling to assess whether there were alterations in the expression of soluble factors or membrane-bound receptors responsible for mediating neuron-glia communication. As an illustration, the neurotransmitters glutamate and acetylcholine can interact with oligodendrocyte receptors, thereby influencing oligodendrocyte proliferation, migration, and differentiation [212,214,215]. As mentioned above, the cAMP/PKA pathway upon PDE4 inhibition can potentiate 4AP and subsequently increase glutamate release from rat prefrontal cortical neurons, offering a possible hypothesis on neuro-glia communication during myelination that could be investigated further [135].

6.3. Summary

cAMP signaling drives oligodendrocyte development and myelination, with PDE4 inhibition shown to enhance differentiation, maturation, and myelination via ERK/CREB and p38 MAPK/CREB pathways (Fig. 5). However, the exact role of PKA in this process remains unclear, although other pro-myelinating treatments are PKA-dependent [216]. Studies using cAMP analogs suggest that enhanced OPC differentiation and myelin gene expression involve PKA/p38 MAPK/CREB and JNK/ATF-2 pathways, again highlighting the importance of the MAPK family in myelination. Yet, crosstalk between cAMP/PKA or cAMP/EPAC and MAPK is not fully elucidated and requires further investigation. Whereas some studies demonstrate a positive role for EPAC in oligodendrocyte differentiation, maturation, and myelination, it has also been postulated as an age-dependent negative regulator of CNS myelination. Similarly, PKA has been found to inhibit myelination at high rolipram concentrations. Consequently, the roles of EPAC and PKA in oligodendrocyte development and myelination are contradictory and require further research.

7. Future perspectives

7.1. Implications in disease

In this review, five pivotal CNS processes were described: neuroinflammation, neuroplasticity, neurotransmission, neuronal survival, and myelination, all of which are influenced by each other and are implicated in neurodegenerative disorders, including AD, PK, MS, and SCI [121,217,218]. Therefore, it is crucial to understand how the distinct findings of PDE4 inhibition and cAMP stimulation in all these separate processes can be translated into therapeutic approaches. For instance, enhanced PKA signaling has been shown to predominantly promote anti-inflammatory responses. However, PKA activation induces growth cone collapse. Consequently, a general PKA-targeting PDE4 inhibitor could reduce inflammatory responses but impair regeneration, making it difficult to predict the outcomes of neurological disorders. As another example, in traumatic brain diseases such as SCI and TBI, firstly targeting the primary inflammation based on PKA activation or p38 MAPK/GSK3β inhibition, followed by boosting neuronal survival, plasticity, and myelination, would be a promising approach. Besides the interplay in downstream pathways, different CNS processes can influence each other. For example, promoting neuronal survival in a specific matter using cAMP-stimulating agents in an environment with demyelination and pro-inflammatory mediators will ultimately result in impaired neuroregeneration. Therefore, further consideration of (cell-type) specific downstream targets is demanded to create a powerful agent that affects either specific processes or tackles similar processes in different cell types to improve neurodegenerative outcomes.

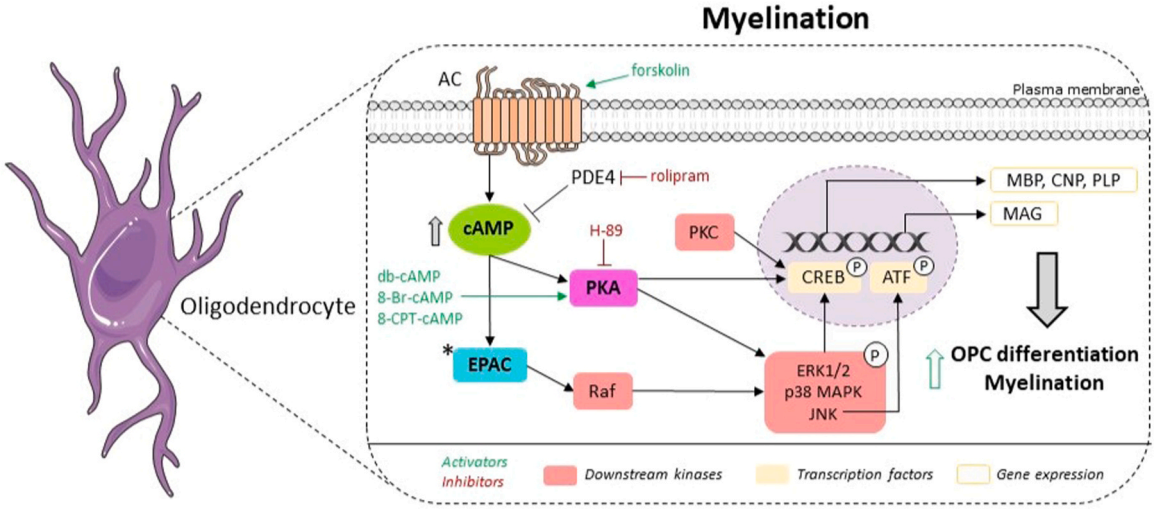

Fig. 5. **:**Downstream signaling upon cAMP stimulation or PDE4 inhibition in myelination**.** Promoting cAMP signaling by inhibiting PDE4 (rolipram) or cAMP analogs specific for PKA (db-cAMP, 8-Br-cAMP, or 8-CPT-cAMPS) and/or EPAC promotes OPC differentiation and myelination. PDE4 inhibition promotes OPC differentiation and myelination by stimulating EPAC/ p38 MAPK signaling, resulting in CREB phosphorylation and MBP, CNP, and PLP gene expression. cAMP analogs promote OPC differentiation and myelination through CREB signaling directly or indirectly via ERK1/2 or p38 MAPK. In addition, PKA/JNK promotes ATF-induced gene expression of MAG to ameliorate OPC differentiation and increase myelination. *AC, Adeylyl cyclase; PDE4, Phosphodiesterase 4; cAMP, Cyclic adenosine monophosphate; PKA, Protein kinase A; db-cAMP, Di-butyryl-cyclic AMP; 8-Br-cAMP, 8-Bromo-cyclic AMP; 8-pCPT, 8-(4-chlorophenylthio)-2′-O-methyl-cyclic AMP; CREB, Cyclic AMP response element binding; MBP, Myelin basic protein; CNP, C-type natriuretic peptide; PLP, Pyridoxal 5′-phosphate; ATF, Activating Transcription Factor; MAG, Myelin-associated glycoprotein; EPAC, Exchange proteins activated directly by cyclic AMP; ERK, Extracellular signal-regulated kinase; p38 MAPK, Mitogen-activated protein kinase; JNK, c-Jun N-terminal kinase; OPC, Oligodendrocyte precursor cell.* *EPAC has also been demonstrated to negatively regulate myelination in an age-dependent manner [211].

7.2. Other PDE inhibitors

PDE4 inhibitors thus far failed to reach the clinic for CNS disorders due to side effects (emesis, nausea, diarrhea, and headaches) at the clinically relevant dose [20,219]. Although ibudilast, roflumilast, and zatolmilast are still in clinical trials for CNS disorders [24,25], finding more specific targets downstream of PDE4 inhibition could offer a new approach to treat neurological disorders. A second option is to focus on gene-specific PDE4 inhibitors (e.g., PDE4D or PDE4B) or isoform-specific PDE4 inhibitors (e.g., PDE4D6) [8,26]. Thirdly, other PDE inhibitors, both cAMP-specific or with dual specificity, that are less well characterized could be investigated further. In particular, PDE7 inhibitors are known not to cause emetic adverse effects and have been shown to promote neuroplasticity, neuronal survival, and OPC differentiation through the cAMP/PKA/CREB pathway [173,220–223]. In addition, PDE7 inhibition can modulate neuroinflammation through inhibition of the MAPK/ERK and NF-κB pathways [59]. Not much research has been conducted on inhibition of PDE8. Currently, there is a limited number of PDE8 inhibitors, and they are not yet under clinical investigation. PDE8A is not expressed in the brain, whereas PDE8B has a restricted expression pattern in the hippocampus, ventral striatum, and cerebellum. Moreover, PDE8B shows increased expression in the brains of AD patients, and its antagonism has been shown to enhance cognition, so the therapeutic potential of PDE8 inhibitors cannot be ruled out [224–226]. In the context of MS, PDE8 inhibition has been shown to reduce neuroinflammation as PDE8 is highly expressed by pro-inflammatory T-cells [227]. The dual-selective PDE1, PDE2, PDE3, PDE10, and PDE11 hydrolyze both cAMP and cGMP; thus, functional outcomes after dual-substrate PDE inhibition should be interpreted carefully since the partly overlapping cGMP signaling may influence experimental outcomes. Inhibiting PDE1 is a widely studied therapeutic approach to enhance neurocognitive functions in treating neurodegenerative disorders, including AD [228]. In addition, PDE3 is being targeted to elevate cAMP levels and, consequently, improve cognition in CNS diseases, as reviewed by *Yanae et al.* [28]. Inhibition of PDE2 and PDE10 is being investigated to treat cognitive-related disorders [229] and schizophrenia and psychosis, respectively [230]. On the contrary, the role of PDE11 in the brain is not well understood yet, and its tissue expression patterns are still under debate [28,231,232].

7.3. cAMP compartmentalization

Over the last 20 years, it has become clear that the cAMP signaling pathway is compartmentalized in subcellular nanodomains within the cell and that spatial confinement and local regulation are critical for the specificity of cAMP signaling and its outcome [233,234]. These distinct nanodomains can explain the opposite roles of PKA and EPAC, two cAMP effectors, in neuronal survival; cAMP elevations have been shown to stimulate neuronal apoptosis through EPAC/p38 MAPK/Bim/Blc2 and PKA/MAPK pathways, whereas it can stimulate neuronal survival through PKA/GSK3/Nrf2 and EPAC/Akt pathways. Consequently, a linear model wherein a message is transduced from GPCR to intracellular effectors through cAMP is too simplistic. Defining the organization of such nanocompartments is crucial to understanding the complex outcomes of pharmacological treatments that elevate cAMP levels. Evidence supporting the model of compartmentalized cAMP signaling is thoroughly reviewed by *Zaccolo et al.*, who also highlights the contribution of various pathway components, including GPCRs, ACs, PKA, AKAPs, EPAC, and phosphoprotein phosphatases, to cAMP compartmentalization, and summarizes experimentally validated cAMP nanosignalomes [234].

cAMP nanodomains can be imaged in real-time using fluorescence resonance energy transfer (FRET)-based cAMP biosensors, which can monitor cAMP dynamics with high spatial and temporal resolution using conventional microscopy [234]. Other genetically encoded cAMP sensors are based on fluorescent proteins or bioluminescence. These methods are thoroughly reviewed elsewhere [235–238]. A recent study has explored cAMP compartments of 60 nm linked to individual GPCRs, called receptor-associated independent cAMP nanodomains (RAINs) [233]. Another study focusing on the role of PDEs in confining cAMP compartments was able to map low-cAMP nanodomains around PDEs ranging between 10 and 30 nm using FRET-cAMP nanoruler technology, suggesting that RAINs may contain multiple PDE nanodomains [239,240]. In addition, it was shown that the PDE-controlled cAMP concentration gradients translated into graded PKA activity, demonstrating that cAMP can elicit local responses that are spatially limited at the nanometer scale [240]. More specifically, research has already revealed some cAMP neuronal compartments that mediate neuronal growth and survival. One study found a cAMP perinuclear neuronal compartment that was largely organized by the multivalent scaffold protein mAKAPα. Moreover, disrupting the PDE4D3-mAKAPα binding promoted mAKAPα-associated cAMP signaling and increased retinal ganglion cell survival *in vivo*. This illustrates a neuronal compartment that regulated cAMP-mediated neuroprotection, offering a novel therapeutic target for CNS indications by inhibiting PDE4D3 [105]. Hence, the ultimate outcome of PKA pathways activated upon cAMP stimulation or PDE inhibition depends on the AKAPs in the cell and their subcellular location. For example, PKA is sequestered to different AKAPs, depending on cell type and subcellular location. *Gao et al.* revealed that cells with high A kinase-interacting protein 1 (AKIP-1), containing PKA, promote NF-κB activation by phosphorylating p65 [51]. In contrast, PKA inhibited NF-κB pathway activation in cells with low AKIP-1 complexes [51]. Numerous neurological processes depend on AKAP-mediated compartmentalization, as extensively summarized by Wild and Dell'Acqua [241].

Despite the promising approaches mentioned, capturing the complexity arising from the multicomponent cAMP signaling system is still challenging. To assemble a cAMP signaling compartment, the signal is initially generated by 30 possible Gs-coupled GPCRs, which are subsequently coupled to 9 different ACs. Short-distance cAMP is then regulated by over 77 isoforms of multiple PDEs, transmitting the signal to various downstream effectors such as PKA or EPAC. PKA itself is controlled by AKAPs, of which more than 43 isoforms exist. Consequently, the many possible combinations among these elements render the experimental characterization of cAMP signaling compartments highly impractical [242]. Computational approaches, as discussed below, could offer valuable insights to overcome this challenge.

Finally, many questions persist concerning cAMP compartmentalization and warrant further investigation. First, the number of cAMP domains in a cell and their molecular composition should be defined. To this end, high single-cell resolution imaging techniques could be employed [234]. Nevertheless, imaging techniques such as FRET to study cAMP compartmentalization are currently being questioned since the presence of fluorescent sensors in nano-sized compartments may alter cAMP measurements by sequestering or interacting with cAMP molecules, lowering their availability. [243]. Secondly, the compartments are hypothesized to be cell-specific and dynamic in terms of time and mobility, adding another layer of complexity. Thirdly, the impact of physical barriers, buffering systems, such as intracellular organelles, and viscosity in impeding cAMP diffusion, in addition to cAMP-degrading PDEs, is poorly understood [238,244]. In conclusion, extensive research is needed to fully grasp cAMP compartmentalization before leveraging it as a therapeutic target.

7.4. Omics-approaches to find new downstream targets

Studies investigating downstream signaling molecules upon cAMP elevations, whether through PDE inhibition or cAMP analogs, frequently employ targeted approaches such as western blot analysis, immunocytochemistry, or ELISA. These studies focus solely on (putative) downstream kinases and their targets rather than taking a more holistic approach and investigating further downstream into the differentially

expressed genes due to altered transcription factor activity. Indeed, using the techniques above, the same proteins, like CREB, have been investigated for years, providing valuable knowledge. However, targeting CREB for CNS indications is impractical because of its ubiquitous expression throughout the body [28]. Moreover, multiple lines of evidence show that targeting PDE4, the most studied PDE for inhibition, might not work in a clinical context since various studies show adverse effects [8,219,245,246]. Therefore, it is crucial to understand which downstream players of the cAMP signaling cascade are key mediators of the positive effects seen upon PDE4 inhibition to identify more specific pharmacological targets for CNS diseases. Omics approaches in specific cell types could help to identify novel, often overlooked, targets.

Data derived from genome and transcriptome studies has limited interpretation because it does not provide direct changes in protein content nor posttranslational modifications like phosphorylation and, therefore, does not fully reflect a biological system. Hence, proteomics has rapidly evolved in the past decade for multiple applications, including identifying complex signaling networks in specific diseases [247,248]. High-throughput proteomic methods, including tissue microarray, protein pathway array, and mass spectrometry, are thoroughly reviewed by *Cui et al.* [248]. A recent study employed an integrated phosphoproteomics approach to identify novel and nonobvious cAMP nanodomains in cardiac myocytes. The authors propose that this strategy has the potential to aid in unraveling the entire landscape of subcellular cAMP compartments in all cell types [249]. The most insight into full-cell biology would come from integrative omics approaches that combine different levels of data from genomics, transcriptomics, (phospho)proteomics, and metabolomics [250]. The main challenge of proteomics and integrative omics approaches is high data variability. Proteomics data are sensitive to various factors, including data preparation models, sample condition and preparation variance, instrument types, and analytical methods. Compared to DNA microarrays or next-generation sequencing methods in genomics, proteomic studies inherently have high levels of data noise, making repeatability impractical and biological interpretation difficult [251]. Furthermore, appropriate data analysis methods for integrative omics are still lacking [250]. Multiple pathway and network analysis tools exist to address challenges concerning proteomics data analysis and biological interpretation of obtained results, which are reviewed by *Wu et al.* [251]. In addition, although discordances between transcriptome-level and proteome-level changes are possible, these approaches often point to alterations in similar pathways, which can then be further analyzed using more targeted techniques [250]. Lastly, computational models could provide biological insight and identify candidate therapeutic targets for intervention [250].

7.5. Computational modeling

As pointed out in this review, the PDE-mediated control of cAMP signaling and its effect on downstream targets is intricate and consists of various feedback mechanisms. A promising approach to gaining a deeper understanding of these mechanisms in neuronal and glial cells is by capturing and analyzing relevant interactions through mathematical models. In the past, these models have provided insight into cAMP signaling in neuronal cells. For instance, a study employing ordinary differential equations (ODEs) revealed the role of Ca^{2+}currents in cAMP-mediated DARPP-32 phosphorylation [252]. As explained earlier, cAMP can activate ERK through different pathways, and exploring these dynamics would be challenging in an *in vitro* setting as it is difficult to observe the relevant signaling changes with current imaging methods. Through mathematical modeling, it was elicited that EPAC activates phosphorylated ERK in a shorter period than PKA, which influences late LTP induction [142]. This finding is significant because ERK and PKA have differential effects on distinct PDE isoforms. Thus, it would be beneficial to extend such computational models to include different PDE isoforms and see their impact on LTP induction dynamics. The model used in the PKA-ERK study captured the spatial and stochastic aspect of cAMP signaling, as compared to previous studies that simplified the process by assuming homogeneity and fast diffusion rates in the cell, allowing to consider both the spatial and temporal aspects of cAMP signaling. Clearly, computational modeling approaches offer additional insights into cAMP dynamics and its effect on downstream pathways. However, only a handful of models analyze the influence of specific PDE inhibitors on downstream signaling of cAMP. For example, an ODE model correctly predicted the effect of combining PDE3 inhibitors and AC agonists on cAMP levels and on selected downstream protein levels of VASP in human platelets. This protein is a downstream target of both PKA and PKC. The model could mimic the effects of cAMP-elevating drugs on VASP phosphorylation, as confirmed by comparing the simulated data to experimental data. Although PDE3 is not cAMP-specific, this study confirms the strength of mathematical models [253]. Shifting the focus to neuronal cells, an ODE model was built to explore the role of ERK in dopamine neurotransmission. The *in silico* results, which focused on the influence of different dopamine concentrations, highlighted the intricate interplay between ERK, PDE4, and PKA. More precisely, it was stated that dopamine-mediated ERK phosphorylation increases PDE4 inhibition, resulting in a higher cAMP level and increased dopamine release. Although this study did not investigate the influence of PDE4 levels, it provided a unique insight into the complexity of cAMP signaling [136]. Another study focused on elucidating the influence of different PDE4 isoforms on PKA and ERK dynamics. It was shown that depending on the presence of a specific regulatory domain, the PDE4 isoform concentration might affect ERK and PKA dynamics differently through a specific feedback loop [113]. Since ERK and PKA have different downstream targets, extending this model could potentially provide relevant insight into the downstream changes caused by fluctuations in PDE isoform abundance. One of the obstacles in creating such dynamic signaling models is the lack of knowledge of kinetic parameters. A possible solution to this problem might be integrating omics data into the model. For instance, using proteomics time-series data collected during experiments to calibrate some of the unknown kinetic parameters in an ODE model resulted in a robust model of MAPK signaling in the context of melanoma treatment. This model is particularly interesting as it introduced the concept of virtual "tagging." In short, the information about the origin of phosphorylation was saved as the virtual phosphorylation cascade progressed, which made it possible to deduce which one of the proteins was responsible for the phosphorylation of the downstream target [254]. Since PKA and ERK might share some downstream targets, it would be interesting to implement such a strategy in the context of cAMP signaling in the CNS. In addition to improved parameterization, future computational models could include the spatial aspects of cAMP dynamics and model the signaling crosstalk more holistically.

8. Conclusion

In summary, the intricate interplay of signaling pathways downstream of cAMP, particularly involving PKA and EPAC, underscores the complexity of cellular responses to PDE4 inhibition and cAMP elevation. All the CNS processes described above show that multiple pathways can converge to achieve the same outcome. This partial redundancy in pathways likely ensures the robustness of these processes, each of which is crucial for proper CNS functioning. While PKA-dependent mechanisms mediate neuronal survival, memory processes, and neurotransmission modulation, findings suggest context-dependent roles influenced by cell type, developmental stage, and signaling crosstalk. Although EPAC-mediated signaling is less investigated than PKA, it has been shown to play a role in key CNS processes upon cAMP elevations. Moreover, the significance of potential crosstalk between pathways and the involvement of different PDE4 isoforms and their spatiotemporal regulation further accentuates the complexity of downstream signaling events. Understanding the consensus PKA and EPAC-dependent

pathways, deciphering the nuances of crosstalk, and elucidating the roles of various PDE4 isoforms are essential for unraveling the redundancy and robustness inherent in cellular responses to cAMP modulation. Additionally, the importance of neuron-glia communication and the dynamic regulation of signaling pathways underscore the need for further investigation to comprehensively elucidate the impact of PDE4 inhibition and cAMP elevation on cellular function in both the healthy and diseased CNS. To conclude, by summarizing existing literature on cAMP signaling upon cAMP elevations and identifying gaps, this review aimed to catalyze further research into PDE4-cAMP mediated pathways to find novel, more specific therapeutic targets for neurodegenerative diseases.

CRediT authorship contribution statement

Jana Van Broeckhoven: Writing – review & editing. **Femke Mussen:** Writing – review & editing, Writing – original draft, Visualization. **Emily Willems:** Writing – review & editing, Writing – original draft, Visualization. **Tim Vanmierlo:** Writing – review & editing, Supervision, Conceptualization. **Melissa Schepers:** Writing – review & editing, Supervision, Conceptualization. **Aurélie Carlier:** Writing – review & editing, Supervision, Conceptualization. **Iga Joanna Skorupska:** Writing – review & editing, Writing – original draft, Visualization, Conceptualization. **Zoë Donders:** Writing – review & editing, Writing – original draft, Visualization, Conceptualization.

Declaration of Competing Interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: T. V. and M.S. have a proprietary interest in selective PDE4D inhibitors for the treatment of demyelinating disorders.

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Appendix

Table A.1
PDE inhibitors, cAMP analogs and compounds that inhibit downstream cAMP effectors.

Modulators	Compound	Target	Agonist	Antagonist
PDE inhibitors	IBMX [255]	PDE, TNF–α		X
	Rolipram [256]	PDE4		X
	Roflupram [55]	PDE4		X
	Roflumilast [257]	PDE4		X
	Piclamilast [258]	PDE4		X
	Ibudilast [259]	PDE, MIF, TLR-4, IL-6, IL-1β, TNF-α		X
	FCPR03 [58]	PDE4		X
	GSK256066 [260]	PDE4		X
	Gebr7b [261]	PDE4D		X
	Gebr32a [27]	PDE4D		X
cAMP analogs [269]	Rp-cAMPS-6-Bnz-cAMP	PKA	X	
	Dioctanoyl-cAMP	PKA	X	
	6-Phe-cAMP	PKA	X	
	8-(4-Chlorophenyl)thio-cAMP	PKA	X	
	8-Br-2′-O-Me-cAMP	EPAC	X	
	8-pCPT-2’-O-Me-cAMP/007-AM	EPAC	X	
	pMe-cAMP	EPAC	X	
	Sp-8-BnT-cAMPS	EPAC	X	
	8-Br-cAMP	PKA, EPAC	X	
	Db-cAMP	PKA, EPAC	X	
	Sp-cAMPS	PKA, EPAC	X	
Modulators of cAMP-related signaling molecules	MDL29,951 [193]	GPCR17	X	
	Forskolin [262]	AC	X	
	KT-5720 [263]	PKA	X	
	H-89 [264]	PKA		X
	(m)-PKI [265]	PKA		X
	Rp-cAMPS	PKA		X
	GF109203 [266]	PKC, PKA	X	
	PMA [267]	PKC	X	
	Chelrythrine [268]	PKC		X
	ESI-05/ESI-09 [269]	EPAC		X
	K-G501 [270]	CREB		X
	LY294002 [271]	PI3K		X
	Wortmannin [271]	PI3K		X
	SB-203580 [272]	P38 MAPK		X
	PD-98059 [273]	MAPK/ERK		X
	U016 [274]	JNK		X
	Compound C [275]	AMPK		X
	EX527 [276]	SIRT1		X

IBMX, 3-isobutyl-1-methylxanthine; TNF–α, Tumor necrosis factor α; MIF, a macrophage migration inhibitory factor; TLR-4; toll-like receptor 4, IL; interleukin, Rp-cAMP, Rp-adenosine 3c′,5c′-cyclic monophosphorothioate triethylammonium salt; 6-Phe-cAFMP, 6-phenyl-cyclic AMP; Sp-8-BnT-cAMPS, 8- Benzylthioadenosine- 3′, 5′- cyclic monophosphorothioate; 8-Br-2′-O-Me-cAMP, 8-bromo-2′O methyl-cyclic AMP; 8-pCPT-2’-O-Me-cAMP, 8-(4-chlorophenylthio)-2′-O-methyl-cyclic AMP; pMe-cAMP, 8-p-methoxyphenylthon-2′-O-methyl-cyclic AMP; Sp-cAMPS, Sp-adenosine 3c′,5c′-cyclic monophosphorothioate triethylammonium salt hydrate; db-cAMP, di-

butyryl-cyclic AMP; 8-Br-cAMP, 8-Bromo-cyclic AMP; PKI, Protein kinase inhibitor; PMA, Phorbol 12-myristate 13-acetate; PKA, Protein kinase A; EPAC, Exchange proteins activated directly by cyclic AMP; CREB, cAMP-response element binding protein; PI3K, Phosphatidylinositol 3-kinase; MAPK/ERK, Mitogen-activated protein kinase/ Extracellular signal-regulated kinase; JNK, Jun N-terminal kinase; AMPK, AMP-activated protein kinase; SIRT, Sirtuin.

References

[1] A.C. Newton, M.D. Bootman, J.D. Scott, Second messengers, Cold Spring Harb. Perspect. Biol. 8 (8) (2016).
[2] K. Yan, et al., The cyclic AMP signaling pathway: exploring targets for successful drug discovery (Review), Mol. Med. Rep. 13 (5) (2016) 3715–3723.
[3] Patra, C., et al., Biochemistry, cAMP, in StatPearls. 2024: Treasure Island (FL).
[4] L.P. Tavares, et al., Blame the signaling: Role of cAMP for the resolution of inflammation, Pharm. Res 159 (2020) 105030.
[5] D. Willoughby, D.M. Cooper, Organization and Ca2+ regulation of adenylyl cyclases in cAMP microdomains, Physiol. Rev. 87 (3) (2007) 965–1010.
[6] K. Devasani, Y. Yao, Expression and functions of adenylyl cyclases in the CNS, Fluids Barriers CNS 19 (1) (2022) 23.
[7] F. Mussen, et al., Unleashing spinal cord repair: the role of cAMP-Specific PDE Inhibition in Attenuating Neuroinflammation and Boosting Regeneration after Traumatic Spinal Cord Injury, Int J. Mol. Sci. 24 (9) (2023).
[8] M. Schepers, et al., Targeting phosphodiesterases-towards a tailor-made approach in multiple sclerosis treatment, Front Immunol. 10 (2019) 1727.
[9] G.S. Baillie, G.S. Tejeda, M.P. Kelly, Therapeutic targeting of 3′,5′-cyclic nucleotide phosphodiesterases: inhibition and beyond, Nat. Rev. Drug Discov. 18 (10) (2019) 770–796.
[10] S. Delhaye, B. Bardoni, Role of phosphodiesterases in the pathophysiology of neurodevelopmental disorders, Mol. Psychiatry 26 (9) (2021) 4570–4582.
[11] M.F. Azevedo, et al., Clinical and molecular genetics of the phosphodiesterases (PDEs), Endocr. Rev. 35 (2) (2014) 195–233.
[12] E. Bollen, et al., Improved long-term memory via enhancing cGMP-PKG signaling requires cAMP-PKA signaling, Neuropsychopharmacology 39 (11) (2014) 2497–2505.
[13] J. Liu, et al., Differential roles of PKA and Epac on the production of cytokines in the endotoxin-stimulated primary cultured microglia, J. Mol. Neurosci. 45 (2) (2011) 186–193.
[14] E.K. Argyrousi, et al., Pro-cognitive effect of upregulating cyclic guanosine monophosphate signalling during memory acquisition or early consolidation is mediated by increased AMPA receptor trafficking, J. Psychopharmacol. 34 (1) (2020) 103–114.
[15] D. Wei, et al., Inhibiting cortical protein kinase A in spinal cord injured rats enhances efficacy of rehabilitative training, Exp. Neurol. 283 (Pt A) (2016) 365–374.
[16] M.V. Podda, et al., Dopamine D1-like receptor activation depolarizes medium spiny neurons of the mouse nucleus accumbens by inhibiting inwardly rectifying K+ currents through a cAMP-dependent protein kinase A-independent mechanism, Neuroscience 167 (3) (2010) 678–690.
[17] D.W. Raible, F.A. McMorris, Oligodendrocyte differentiation and progenitor cell proliferation are independently regulated by cyclic AMP, J. Neurosci. Res. 34 (3) (1993) 287–294.
[18] D.J. Propper, et al., Phase I study of the novel cyclic AMP (cAMP) analogue 8-chloro-cAMP in patients with cancer: toxicity, hormonal, and immunological effects, Clin. Cancer Res. 5 (7) (1999) 1682–1689.
[19] E.G. Cameron, M.S. Kapiloff, Intracellular compartmentation of cAMP promotes neuroprotection and regeneration of CNS neurons, Neural Regen. Res. 12 (2) (2017) 201–202.
[20] A. Blokland, et al., Phosphodiesterase Type 4 Inhibition in CNS Diseases, Trends Pharm. Sci. 40 (12) (2019) 971–985.
[21] L.J.S. Sugin, et al., Roflumilast: a potential drug for the treatment of cognitive impairment? Neurosci. Lett. 736 (2020) 135281.
[22] G. Oliveros, et al., Repurposing ibudilast to mitigate Alzheimer's disease by targeting inflammation, Brain 146 (3) (2023) 898–911.
[23] A.D. Bondarev, et al., Recent developments of phosphodiesterase inhibitors: clinical trials, emerging indications and novel molecules, Front Pharm. 13 (2022) 1057083.
[24] L. Crocetti, et al., An overview of PDE4 inhibitors in clinical trials: 2010 to early 2022, Molecules 27 (15) (2022).
[25] G. Li, et al., Advances in the development of phosphodiesterase-4 inhibitors, Eur. J. Med. Chem. 250 (2023) 115195.
[26] M. Schepers, et al., Selective PDE4 subtype inhibition provides new opportunities to intervene in neuroinflammatory versus myelin damaging hallmarks of multiple sclerosis, Brain Behav. Immun. 109 (2023) 1–22.
[27] R. Ricciarelli, et al., Memory-enhancing effects of GEBR-32a, a new PDE4D inhibitor holding promise for the treatment of Alzheimer's disease, Sci. Rep. 7 (2017) 46320.
[28] S. Yanai, S. Endo, PDE3 inhibitors repurposed as treatments for age-related cognitive impairment, Mol. Neurobiol. 56 (6) (2019) 4306–4316.
[29] H. Shiga, H. Asou, E. Ito, Advancement of differentiation of oligodendrocyte progenitor cells by a cascade including protein kinase A and cyclic AMP-response element binding protein, Neurosci. Res 53 (4) (2005) 436–441.
[30] Y. Xu, et al., Inhibition of phosphodiesterase 2 reverses impaired cognition and neuronal remodeling caused by chronic stress, Neurobiol. Aging 36 (2) (2015) 955–970.
[31] V.K. Raker, C. Becker, K. Steinbrink, The cAMP pathway as therapeutic target in autoimmune and inflammatory diseases, Front Immunol. 7 (2016) 123.
[32] N.R. Bhat, P. Zhang, S.B. Mohanty, p38 MAP kinase regulation of oligodendrocyte differentiation with CREB as a potential target, Neurochem. Res. 32 (2) (2007) 293–302.
[33] F. Christian, et al., Small molecule AKAP-protein kinase A (PKA) interaction disruptors that activate PKA interfere with compartmentalized cAMP signaling in cardiac myocytes, J. Biol. Chem. 286 (11) (2011) 9079–9096.
[34] G. Borland, B.O. Smith, S.J. Yarwood, EPAC proteins transduce diverse cellular actions of cAMP, Br. J. Pharm. 158 (1) (2009) 70–86.
[35] H. Tozaki-Saitoh, et al., Involvement of exchange protein directly activated by cAMP and tumor progression locus 2 in IL-1beta production in microglial cells following activation of beta-adrenergic receptors, J. Pharm. Sci. 143 (3) (2020) 133–140.
[36] M. Swire, et al., Oligodendrocyte HCN2 channels regulate myelin sheath length, J. Neurosci. 41 (38) (2021) 7954–7964.
[37] S. Bolton, A.M. Butt, Cyclic AMP-mediated regulation of the resting membrane potential in myelin-forming oligodendrocytes in the isolated intact rat optic nerve, Exp. Neurol. 202 (1) (2006) 36–43.
[38] S. Amor, et al., Inflammation in neurodegenerative diseases, Immunology 129 (2) (2010) 154–169.
[39] H.S. Kwon, S.H. Koh, Neuroinflammation in neurodegenerative disorders: the roles of microglia and astrocytes, Transl. Neurodegener. 9 (1) (2020) 42.
[40] L. Ponsaerts, et al., Neuroinflammation in ischemic stroke: inhibition of cAMP-specific phosphodiesterases (PDEs) to the rescue, Biomedicines 9 (7) (2021).
[41] M. Fakhoury, Microglia and astrocytes in Alzheimer's disease: implications for therapy, Curr. Neuropharmacol. 16 (5) (2018) 508–518.
[42] I.D. Vainchtein, A.V. Molofsky, Astrocytes and microglia: in sickness and in health, Trends Neurosci. 43 (3) (2020) 144–154.
[43] E. Traiffort, et al., Astrocytes and microglia as major players of myelin production in normal and pathological conditions, Front Cell Neurosci. 14 (2020) 79.
[44] M. Lombardi, et al., Detrimental and protective action of microglial extracellular vesicles on myelin lesions: astrocyte involvement in remyelination failure, Acta Neuropathol. 138 (6) (2019) 987–1012.
[45] S.H. Christiansen, et al., Combined anti-inflammatory effects of beta2-adrenergic agonists and PDE4 inhibitors on astrocytes by upregulation of intracellular cAMP, Neurochem Int. 59 (6) (2011) 837–846.
[46] S.A. Myers, et al., Following spinal cord injury, PDE4B drives an acute, local inflammatory response and a chronic, systemic response exacerbated by gut dysbiosis and endotoxemia, Neurobiol. Dis. 124 (2019) 353–363.
[47] M. Ghosh, et al., Proinflammatory cytokine regulation of cyclic AMP-phosphodiesterase 4 signaling in microglia in vitro and following CNS injury, Glia 60 (12) (2012) 1839–1859.
[48] H. Cheng, et al., siRNA-mediated silencing of phosphodiesterase 4B expression affects the production of cytokines in endotoxin-stimulated primary cultured microglia, Exp. Ther. Med. 12 (4) (2016) 2257–2264.
[49] T. Liu, et al., NF-kappaB signaling in inflammation, Signal Transduct. Target Ther. 2 (2017) 17023.
[50] M. Ghosh, et al., The interplay between cyclic AMP, MAPK, and NF-kappaB pathways in response to proinflammatory signals in microglia, Biomed. Res. Int. 2015 (2015) 308461.
[51] N. Gao, et al., A-kinase-interacting protein 1 (AKIP1) acts as a molecular determinant of PKA in NF-kappaB signaling, J. Biol. Chem. 285 (36) (2010) 28097–28104.
[52] F. Christian, E.L. Smith, R.J. Carmody, The regulation of NF-kappaB Subunits by Phosphorylation, Cells 5 (1) (2016).
[53] H. Guo, et al., FFPM, a PDE4 inhibitor, reverses learning and memory deficits in APP/PS1 transgenic mice via cAMP/PKA/CREB signaling and anti-inflammatory effects, Neuropharmacology 116 (2017) 260–269.
[54] D.Y. Kim, et al., The potent PDE10A inhibitor MP-10 (PF-2545920) suppresses microglial activation in LPS-induced neuroinflammation and MPTP-induced Parkinson's disease mouse models, J. Neuroimmune Pharm. 16 (2) (2021) 470–482.
[55] D. Li, et al., Roflupram, a novel phosphodiesterase 4 inhibitor, inhibits lipopolysaccharide-induced neuroinflammatory responses through activation of the AMPK/Sirt1 pathway, Int Immunopharmacol. 90 (2021) 107176.
[56] D.Y. Xia, et al., SIRT1 Promotes M2 microglia polarization via reducing ROS-mediated NLRP3 inflammasome signaling after subarachnoid hemorrhage, Front Immunol. 12 (2021) 770744.
[57] W. Zhang, et al., SIRT1 inactivation switches reactive astrocytes to an antiinflammatory phenotype in CNS autoimmunity, J. Clin. Invest (22) (2022) **132**.
[58] Z.Q. Zou, et al., Novel phosphodiesterase 4 inhibitor FCPR03 alleviates lipopolysaccharide-induced neuroinflammation by regulation of the cAMP/PKA/CREB signaling pathway and NF-kappaB inhibition, J. Pharm. Exp. Ther. 362 (1) (2017) 67–77.
[59] I. Paterniti, et al., PDE 7 inhibitors: new potential drugs for the therapy of spinal cord injury, PLoS One 6 (1) (2011) e15937.

[60] H.Y. Wu, et al., Both classic Gs-cAMP/PKA/CREB and alternative Gs-cAMP/PKA/p38beta/CREB signal pathways mediate exenatide-stimulated expression of M2 microglial markers. J. Neuroimmunol. 316 (2018) 17–22.
[61] M. Ghosh, Y. Xu, D.D. Pearse, Cyclic AMP is a key regulator of M1 to M2a phenotypic conversion of microglia in the presence of Th2 cytokines, J. Neuroinflamm. 13 (2016) 9.
[62] A. Guijarro-Belmar, et al., Epac2 elevation reverses inhibition by chondroitin sulfate proteoglycans in vitro and transforms postlesion inhibitory environment to promote axonal outgrowth in an Ex vivo model of spinal cord injury, J. Neurosci. 39 (42) (2019) 8330–8346.
[63] O. Adolph, et al., Rapid increase of glial glutamate uptake via blockade of the protein kinase A pathway, Glia 55 (16) (2007) 1699–1707.
[64] B. Draganski, A. May, Training-induced structural changes in the adult human brain, Behav. Brain Res. 192 (1) (2008) 137–142.
[65] R. Barkhordar, et al., Reducing mercury vapor, CDA J. 15 (11) (1987) 57–59.
[66] B. Leuner, E. Gould, Structural plasticity and hippocampal function, Annu Rev. Psychol. 61 (2010) 111–140. C1-3.
[67] X. Chen, et al., Neonatal exposure to low-dose (1.2%) sevoflurane increases rats' hippocampal neurogenesis and synaptic plasticity in later life, Neurotox. Res. 34 (2) (2018) 188–197.
[68] G. Kempermann, H. Song, F.H. Gage, Neurogenesis in the adult hippocampus, Cold Spring Harb. Perspect. Biol. 7 (9) (2015) a018812.
[69] K.L. Hedstrom, Y. Ogawa, M.N. Rasband, AnkyrinG is required for maintenance of the axon initial segment and neuronal polarity, J. Cell Biol. 183 (4) (2008) 635–640.
[70] S. Gomis-Ruth, C.J. Wierenga, F. Bradke, Plasticity of polarization: changing dendrites into axons in neurons integrated in neuronal circuits, Curr. Biol. 18 (13) (2008) 992–1000.
[71] A. Citri, R.C. Malenka, Synaptic plasticity: multiple forms, functions, and mechanisms, Neuropsychopharmacology 33 (1) (2008) 18–41.
[72] B. Rombaut, et al., PDE inhibition in distinct cell types to reclaim the balance of synaptic plasticity, Theranostics 11 (5) (2021) 2080–2097.
[73] G.M. Petzinger, et al., Exercise-enhanced neuroplasticity targeting motor and cognitive circuitry in Parkinson's disease, Lancet Neurol. 12 (7) (2013) 716–726.
[74] Y. Ding, A.J. Kastin, W. Pan, Neural plasticity after spinal cord injury, Curr. Pharm. Des. 11 (11) (2005) 1441–1450.
[75] M.A. Dimyan, L.G. Cohen, Neuroplasticity in the context of motor rehabilitation after stroke, Nat. Rev. Neurol. 7 (2) (2011) 76–85.
[76] H. Feng, et al., Roflumilast ameliorates cognitive impairment in APP/PS1 mice via cAMP/CREB/BDNF signaling and anti-neuroinflammatory effects, Metab. Brain Dis. 34 (2) (2019) 583–591.
[77] H. Wang, et al., cAMP response element-binding protein (CREB): a possible signaling molecule link in the pathophysiology of schizophrenia, Front Mol. Neurosci. 11 (2018) 255.
[78] Y. Wang, et al., TrkB/BDNF signaling pathway and its small molecular agonists in CNS injury, Life Sci. 336 (2024) 122282.
[79] T.R. Ismail, et al., Environmental enrichment and metformin improve metabolic functions, hippocampal neuron survival, and hippocampal-dependent memory in high-fat/high-sucrose diet-induced type 2 diabetic rats, Biology 12 (3) (2023).
[80] J. Ster, et al., Epac mediates PACAP-dependent long-term depression in the hippocampus, J. Physiol. 587 (1) (2009) 101–113.
[81] P. Penzes, K.M. Woolfrey, D.P. Srivastava, Epac2-mediated dendritic spine remodeling: implications for disease, Mol. Cell Neurosci. 46 (2) (2011) 368–380.
[82] A.J. Murray, D.A. Shewan, Epac mediates cyclic AMP-dependent axon growth, guidance and regeneration, Mol. Cell Neurosci. 38 (4) (2008) 578–588.
[83] S.L. Mironov, E.Y. Skorova, S. Kugler, Epac-mediated cAMP-signalling in the mouse model of Rett Syndrome, Neuropharmacology 60 (6) (2011) 869–877.
[84] S.D. Boomkamp, et al., Epac and the high affinity rolipram binding conformer of PDE4 modulate neurite outgrowth and myelination using an in vitro spinal cord injury model, Br. J. Pharm. 171 (9) (2014) 2385–2398.
[85] N. Hasan, et al., Roflumilast reduces pathological symptoms of sporadic Alzheimer's disease in rats produced by intracerebroventricular streptozotocin by inhibiting NF-kappaB/BACE-1 mediated abeta production in the hippocampus and activating the cAMP/BDNF signalling pathway. Neurotox. Res. 40 (2) (2022) 432–448.
[86] H.A. Farid, et al., PI3K/AKT signaling activation by roflumilast ameliorates rotenone-induced Parkinson's disease in rats, Inflammopharmacology (2023).
[87] S.S. Hannila, M.T. Filbin, The role of cyclic AMP signaling in promoting axonal regeneration after spinal cord injury, Exp. Neurol. 209 (2) (2008) 321–332.
[88] Y.M. Fleming, M.C. Frame, M.D. Houslay, PDE4-regulated cAMP degradation controls the assembly of integrin-dependent actin adhesion structures and REF52 cell migration, J. Cell Sci. 117 (Pt 11) (2004) 2377–2388.
[89] F. Drees, F.B. Gertler, Ena/VASP: proteins at the tip of the nervous system, Curr. Opin. Neurobiol. 18 (1) (2008) 53–59.
[90] A.S. Sierksma, et al., Improvement of spatial memory function in APPswe/PS1dE9 mice after chronic inhibition of phosphodiesterase type 4D, Neuropharmacology 77 (2014) 120–130.
[91] D. Paes, et al., Ablation of specific long PDE4D isoforms increases neurite elongation and conveys protection against amyloid-beta pathology, Cell Mol. Life Sci. 80 (7) (2023) 178.
[92] J. Schwenkgrub, et al., The phosphodiesterase inhibitor, ibudilast, attenuates neuroinflammation in the MPTP model of Parkinson's disease, PLoS One 12 (7) (2017) e0182019.
[93] T. Mizuno, et al., Neuroprotective role of phosphodiesterase inhibitor ibudilast on neuronal cell death induced by activated microglia, Neuropharmacology 46 (3) (2004) 404–411.
[94] R. Gopalakrishna, et al., Cyclic-AMP induces Nogo-A receptor NgR1 internalization and inhibits Nogo-A-mediated collapse of growth cone, Biochem Biophys. Res. Commun. 523 (3) (2020) 678–684.
[95] G.F. Grinstead, A closer look at acetyl and pentafluoropropionyl derivatives for quantitative analysis of morphine and codeine by gas chromatography/mass spectrometry, J. Anal. Toxicol. 15 (6) (1991) 293–298.
[96] A.W. McGee, S.M. Strittmatter, The Nogo-66 receptor: focusing myelin inhibition of axon regeneration, Trends Neurosci. 26 (4) (2003) 193–198.
[97] M. Lehmann, et al., Inactivation of Rho signaling pathway promotes CNS axon regeneration, J. Neurosci. 19 (17) (1999) 7537–7547.
[98] N. Forgione, M.G. Fehlings, Rho-ROCK inhibition in the treatment of spinal cord injury, World Neurosurg. 82 (3-4) (2014) e535–e539.
[99] M.C. Renner, et al., Synaptic plasticity through activation of GluA3-containing AMPA-receptors, Elife 6 (2017).
[100] S.H. Shi, et al., Rapid spine delivery and redistribution of AMPA receptors after synaptic NMDA receptor activation, Science 284 (5421) (1999) 1811–1816.
[101] S. Shi, et al., Subunit-specific rules governing AMPA receptor trafficking to synapses in hippocampal pyramidal neurons, Cell 105 (3) (2001) 331–343.
[102] Y. Hayashi, et al., Driving AMPA receptors into synapses by LTP and CaMKII: requirement for GluR1 and PDZ domain interaction, Science 287 (5461) (2000) 2262–2267.
[103] S.L. Campbell, et al., Altered phosphorylation, electrophysiology, and behavior on attenuation of PDE4B action in hippocampus, BMC Neurosci. 18 (1) (2017) 77.
[104] D. Paes, et al., Increased isoform-specific phosphodiesterase 4D expression is associated with pathology and cognitive impairment in Alzheimer's disease, Neurobiol. Aging 97 (2021) 56–64.
[105] T. Boczek, et al., Regulation of neuronal survival and axon growth by a perinuclear cAMP compartment, J. Neurosci. 39 (28) (2019) 5466–5480.
[106] V. De Arcangelis, et al., Differential association of phosphodiesterase 4D isoforms with beta2-adrenoceptor in cardiac myocytes, J. Biol. Chem. 284 (49) (2009) 33824–33832.
[107] G.S. Chai, et al., Activation of beta(2)-adrenergic receptor promotes dendrite ramification and spine generation in APP/PS1 mice, Neurosci. Lett. 636 (2017) 158–164.
[108] M.P. Mattson, A. Taylor-Hunter, S.B. Kater, Neurite outgrowth in individual neurons of a neuronal population is differentially regulated by calcium and cyclic AMP, J. Neurosci. 8 (5) (1988) 1704–1711.
[109] S. Sanchez, et al., A cAMP-activated pathway, including PKA and PI3K, regulates neuronal differentiation, Neurochem Int 44 (4) (2004) 231–242.
[110] N. Nakao, An increase in intracellular levels of cyclic AMP produces trophic effects on striatal neurons developing in culture, Neuroscience 82 (4) (1998) 1009–1020.
[111] D. Cai, et al., Arginase I and polyamines act downstream from cyclic AMP in overcoming inhibition of axonal growth MAG and myelin in vitro, Neuron 35 (4) (2002) 711–719.
[112] A.C. Emery, L.E. Eiden, Signaling through the neuropeptide GPCR PAC(1) induces neuritogenesis via a single linear cAMP- and ERK-dependent pathway using a novel cAMP sensor, FASEB J. 26 (8) (2012) 3199–3211.
[113] D. Paes, et al., Computational investigation of the dynamic control of cAMP signaling by PDE4 isoform types, Biophys. J. 121 (14) (2022) 2693–2711.
[114] S. Ojea Ramos, M. Feld, M.S. Fustinana, Contributions of extracellular-signal regulated kinase 1/2 activity to the memory trace, Front Mol. Neurosci. 15 (2022) 988790.
[115] M. Kostic, N. Zivkovic, I. Stojanovic, Multiple sclerosis and glutamate excitotoxicity, Rev. Neurosci. 24 (1) (2013) 71–88.
[116] C. Rangel-Barajas, I. Coronel, B. Floran, Dopamine receptors and neurodegeneration, Aging Dis. 6 (5) (2015) 349–368.
[117] J. Segura-Aguilar, et al., Protective and toxic roles of dopamine in Parkinson's disease, J. Neurochem 129 (6) (2014) 898–915.
[118] A. Sood, et al., Glia: A major player in glutamate-GABA dysregulation-mediated neurodegeneration, J. Neurosci. Res. 99 (12) (2021) 3148–3189.
[119] S.G. Snowden, et al., Neurotransmitter imbalance in the brain and Alzheimer's disease pathology, J. Alzheimers Dis. 72 (1) (2019) 35–43.
[120] G. Koch, et al., Impaired LTP- but not LTD-like cortical plasticity in Alzheimer's disease patients, J. Alzheimers Dis. 31 (3) (2012) 593–599.
[121] E. Akyuz, et al., Exploring the role of neurotransmitters in multiple sclerosis: an expanded review, ACS Chem. Neurosci. 14 (4) (2023) 527–553.
[122] S. Weiss, et al., Disability in multiple sclerosis: when synaptic long-term potentiation fails, Neurosci. Biobehav Rev. 43 (2014) 88–99.
[123] P. Greengard, et al., Enhancement of the glutamate response by cAMP-dependent protein kinase in hippocampal neurons, Science 253 (5024) (1991) 1135–1138.
[124] K. Obrietan, A.N. van den Pol, GABA activity mediating cytosolic Ca2+ rises in developing neurons is modulated by cAMP-dependent signal transduction, J. Neurosci. 17 (12) (1997) 4785–4799.
[125] P. Svenningsson, et al., DARPP-32: an integrator of neurotransmission, Annu Rev. Pharm. Toxicol. 44 (2004) 269–296.
[126] M.A. Lynch, Long-term potentiation and memory, Physiol. Rev. 84 (1) (2004) 87–136.
[127] R.D. Blitzer, et al., Postsynaptic cAMP pathway gates early LTP in hippocampal CA1 region, Neuron 15 (6) (1995) 1403–1414.
[128] M.E. Bach, et al., Age-related defects in spatial memory are correlated with defects in the late phase of hippocampal long-term potentiation in vitro and are attenuated by drugs that enhance the cAMP signaling pathway, Proc. Natl. Acad. Sci. 96 (9) (1999) 5280–5285.
[129] O.A. Reneerkens, et al., Selective phosphodiesterase inhibitors: a promising target for cognition enhancement, Psychopharmacol. 202 (1-3) (2009) 419–443.

[130] E. Mota, et al., Pivotal role of phosphodiesterase 10A in the integration of dopamine signals in mice striatal D(1) and D(2) medium-sized spiny neurones, Br. J. Pharm. 178 (24) (2021) 4873–4890.
[131] P. Greengard, P.B. Allen, A.C. Nairn, Beyond the dopamine receptor: the DARPP-32/protein phosphatase-1 cascade, Neuron 23 (3) (1999) 435–447.
[132] A. Nishi, et al., Distinct roles of PDE4 and PDE10A in the regulation of cAMP/PKA signaling in the striatum, J. Neurosci. 28 (42) (2008) 10460–10471.
[133] M. Kuroiwa, et al., Phosphodiesterase 4 inhibition enhances the dopamine D1 receptor/PKA/DARPP-32 signaling cascade in frontal cortex, Psychopharmacol. 219 (4) (2012) 1065–1079.
[134] H. Bito, K. Deisseroth, R.W. Tsien, CREB phosphorylation and dephosphorylation: a Ca(2+)- and stimulus duration-dependent switch for hippocampal gene expression, Cell 87 (7) (1996) 1203–1214.
[135] S.J. Wang, An investigation into the effect of the type IV phosphodiesterase inhibitor rolipram in the modulation of glutamate release from rat prefrontocortical nerve terminals, Synapse 59 (1) (2006) 41–50.
[136] R.S. Song, et al., ERK regulation of phosphodiesterase 4 enhances dopamine-stimulated AMPA receptor membrane insertion, Proc. Natl. Acad. Sci. 110 (38) (2013) 15437–15442.
[137] S. Ryan, et al., Repeated shock stress facilitates basolateral amygdala synaptic plasticity through decreased cAMP-specific phosphodiesterase type IV (PDE4) expression, Brain Struct. Funct. 223 (4) (2018) 1731–1745.
[138] T. Ahmed, J.U. Frey, Expression of the specific type IV phosphodiesterase gene PDE4B3 during different phases of long-term potentiation in single hippocampal slices of rats in vitro, Neuroscience 117 (3) (2003) 627–638.
[139] M. Barad, et al., Rolipram, a type IV-specific phosphodiesterase inhibitor, facilitates the establishment of long-lasting long-term potentiation and improves memory, Proc. Natl. Acad. Sci. 95 (25) (1998) 15020–15025.
[140] K. Rutten, et al., Enhanced long-term potentiation and impaired learning in phosphodiesterase 4D-knockout (PDE4D) mice, Eur. J. Neurosci. 28 (3) (2008) 625–632.
[141] V. Wiescholleck, D. Manahan-Vaughan, PDE4 inhibition enhances hippocampal synaptic plasticity in vivo and rescues MK801-induced impairment of long-term potentiation and object recognition memory in an animal model of psychosis, Transl. Psychiatry 2 (3) (2012) e89.
[142] N.T. Miningou Zobon, J. Jedrzejewska-Szmek, K.T. Blackwell, Temporal pattern and synergy influence activity of ERK signaling pathways during L-LTP induction, Elife 10 (2021).
[143] S.X. Jin, et al., Abeta oligomers from human brain impair mossy fiber LTP in CA3 of hippocampus, but activating cAMP-PKA and cGMP-PKG prevents this, Neurobiol. Dis. 172 (2022) 105816.
[144] J. Li, et al., Dopamine D2-like antagonists induce chromatin remodeling in striatal neurons through cyclic AMP-protein kinase A and NMDA receptor signaling, J. Neurochem 90 (5) (2004) 1117–1131.
[145] W.X. Shi, B.S. Bunney, Roles of intracellular cAMP and protein kinase A in the actions of dopamine and neurotensin on midbrain dopamine neurons, J. Neurosci. 12 (6) (1992) 2433–2438.
[146] G. Page, et al., The up-regulation of the striatal dopamine transporter's activity by cAMP is PKA-, CaMK II- and phosphatase-dependent. Neurochem Int 45 (5) (2004) 627–632.
[147] I. Herrero, J. Sanchez-Prieto, cAMP-dependent facilitation of glutamate release by beta-adrenergic receptors in cerebrocortical nerve terminals, J. Biol. Chem. 271 (48) (1996) 30554–30560.
[148] J.J. Ferrero, et al., beta-Adrenergic receptors activate exchange protein directly activated by cAMP (Epac), translocate Munc13-1, and enhance the Rab3A-RIM1alpha interaction to potentiate glutamate release at cerebrocortical nerve terminals, J. Biol. Chem. 288 (43) (2013) 31370–31385.
[149] G.E. Gochenauer, M.B. Robinson, Dibutyryl-cAMP (dbcAMP) up-regulates astrocytic chloride-dependent L-[3H]glutamate transport and expression of both system xc(-) subunits, J. Neurochem 78 (2) (2001) 276–286.
[150] B.D. Schlag, et al., Regulation of the glial Na+-dependent glutamate transporters by cyclic AMP analogs and neurons, Mol. Pharm. 53 (3) (1998) 355–369.
[151] H.R. Zielke, et al., Effect of 8-bromo-cAMP and dexamethasone on glutamate metabolism in rat astrocytes, Neurochem Res. 15 (11) (1990) 1115–1122.
[152] M.H. Tehrani, J.J. Hablitz, E.M. Barnes Jr., cAMP increases the rate of GABAA receptor desensitization in chick cortical neurons, Synapse 4 (2) (1989) 126–131.
[153] S.J. Moss, et al., Functional modulation of GABAA receptors by cAMP-dependent protein phosphorylation, Science 257 (5070) (1992) 661–665.
[154] G. Heuschneider, R.D. Schwartz, cAMP and forskolin decrease gamma-aminobutyric acid-gated chloride flux in rat brain synaptoneurosomes, Proc. Natl. Acad. Sci. 86 (8) (1989) 2938–2942.
[155] A. Ganguly, D. Lee, Suppression of inhibitory GABAergic transmission by cAMP signaling pathway: alterations in learning and memory mutants, Eur. J. Neurosci. 37 (9) (2013) 1383–1393.
[156] F. Niwa, et al., cAMP-EPAC-dependent regulation of gephyrin phosphorylation and GABA(A)R trapping at inhibitory synapses. iScience 22 (2019) 453–465.
[157] T. Bartsch, et al., CA1 neurons in the human hippocampus are critical for autobiographical memory, mental time travel, and autonoetic consciousness, Proc. Natl. Acad. Sci. 108 (42) (2011) 17562–17567.
[158] U. Frey, Y.Y. Huang, E.R. Kandel, Effects of cAMP simulate a late stage of LTP in hippocampal CA1 neurons, Science 260 (5114) (1993) 1661–1664.
[159] J.R. Slack, C. Walsh, Effects of a cAMP analogue simulate the distinct components of long-term potentiation in CA1 region of rat hippocampus, Neurosci. Lett. 201 (1) (1995) 25–28.
[160] S.T. Wong, et al., Calcium-stimulated adenylyl cyclase activity is critical for hippocampus-dependent long-term memory and late phase LTP, Neuron 23 (4) (1999) 787–798.
[161] B.I. Kanterewicz, et al., The extracellular signal-regulated kinase cascade is required for NMDA receptor-independent LTP in area CA1 but not area CA3 of the hippocampus, J. Neurosci. 20 (9) (2000) 3057–3066.
[162] N. Otmakhov, et al., Forskolin-induced LTP in the CA1 hippocampal region is NMDA receptor dependent, J. Neurophysiol. 91 (5) (2004) 1955–1962.
[163] C. Wozny, et al., Differential cAMP signaling at hippocampal output synapses, J. Neurosci. 28 (53) (2008) 14358–14362.
[164] Q.W. Wang, M.J. Rowan, R. Anwyl, Inhibition of LTP by beta-amyloid is prevented by activation of beta2 adrenoceptors and stimulation of the cAMP/PKA signalling pathway, Neurobiol. Aging 30 (10) (2009) 1608–1613.
[165] O.V. Vitolo, et al., Amyloid beta -peptide inhibition of the PKA/CREB pathway and long-term potentiation: reversibility by drugs that enhance cAMP signaling, Proc. Natl. Acad. Sci. 99 (20) (2002) 13217–13221.
[166] J.N. Gelinas, et al., Activation of exchange protein activated by cyclic-AMP enhances long-lasting synaptic potentiation in the hippocampus, Learn Mem. 15 (6) (2008) 403–411.
[167] B.J. Andreone, M. Larhammar, J.W. Lewcock, Cell death and neurodegeneration, Cold Spring Harb. Perspect. Biol. 12 (2) (2020).
[168] R. Donev, et al., Neuronal death in Alzheimer's disease and therapeutic opportunities, J. Cell Mol. Med 13 (11-12) (2009) 4329–4348.
[169] M.J. Benskey, R.G. Perez, F.P. Manfredsson, The contribution of alpha synuclein to neuronal survival and function - Implications for Parkinson's disease, J. Neurochem 137 (3) (2016) 331–359.
[170] S.A. Johnson, et al., Pharmacotherapy for amyotrophic lateral sclerosis: a review of approved and upcoming agents, Drugs 82 (13) (2022) 1367–1388.
[171] B. Benedetti, et al., Spinal cord injury and loss of cortical inhibition, Int. J. Mol. Sci. 23 (10) (2022).
[172] Y.F. Cong, et al., Rolipram ameliorates memory deficits and depression-like behavior in APP/PS1/tau triple transgenic mice: involvement of neuroinflammation and apoptosis via cAMP signaling, Int. J. Neuropsychopharmacol. 26 (9) (2023) 585–598.
[173] J.A. Morales-Garcia, et al., Phosphodiesterase 7 inhibition preserves dopaminergic neurons in cellular and rodent models of Parkinson disease, PLoS One 6 (2) (2011) e17240.
[174] B. Zeng, et al., Involvement of PI3K/Akt/FoxO3a and PKA/CREB signaling pathways in the protective effect of fluoxetine against corticosterone-induced cytotoxicity in PC12 Cells, J. Mol. Neurosci. 59 (4) (2016) 567–578.
[175] M.N. Lague, et al., Synergistic effects of Pten loss and WNT/CTNNB1 signaling pathway activation in ovarian granulosa cell tumor development and progression, Carcinogenesis 29 (11) (2008) 2062–2072.
[176] B. Xu, et al., Roflumilast prevents ischemic stroke-induced neuronal damage by restricting GSK3beta-mediated oxidative stress and IRE1alpha/TRAF2/JNK pathway. Free Radic. Biol. Med. 163 (2021) 281–296.
[177] K. Lee, Epac: new emerging cAMP-binding protein, BMB Rep. 54 (3) (2021) 149–156.
[178] A. Merighi, L. Lossi, Endoplasmic reticulum stress signaling and neuronal cell death, Int. J. Mol. Sci. 23 (23) (2022).
[179] J. Xiao, et al., Inhibition of PDE4 Attenuates TNF-alpha-triggered cell death through suppressing NF-kappaB and JNK activation in HT-22 neuronal cells. Cell Mol. Neurobiol. 40 (3) (2020) 421–435.
[180] J. Zhong, et al., Roflupram exerts neuroprotection via activation of CREB/PGC-1alpha signalling in experimental models of Parkinson's disease, Br. J. Pharm. 177 (10) (2020) 2333–2350.
[181] Z. Kuczynska, et al., Covering the role of PGC-1alpha in the nervous system, Cells 11 (1) (2021).
[182] T. Sasaki, et al., The phosphodiesterase inhibitor rolipram promotes survival of newborn hippocampal neurons after ischemia, Stroke 38 (5) (2007) 1597–1605.
[183] K. Kranz, et al., Phosphodiesterase type 4 inhibitor rolipram improves survival of spiral ganglion neurons in vitro, PLoS One 9 (3) (2014) e92157.
[184] M. Li, et al., Cyclic AMP promotes neuronal survival by phosphorylation of glycogen synthase kinase 3beta, Mol. Cell Biol. 20 (24) (2000) 9356–9363.
[185] R. Guan, et al., Potential role of the cAMP/PKA/CREB signalling pathway in hypoxic preconditioning and effect on propofol-induced neurotoxicity in the hippocampus of neonatal rats. Mol. Med. Rep. 20 (2) (2019) 1837–1845.
[186] C. Aglah, T. Gordon, E.I. Posse de Chaves, cAMP promotes neurite outgrowth and extension through protein kinase A but independently of Erk activation in cultured rat motoneurons, Neuropharmacology 55 (1) (2008) 8–17.
[187] S. Poser, et al., Brain-derived neurotrophic factor protection of cortical neurons from serum withdrawal-induced apoptosis is inhibited by cAMP, J. Neurosci. 23 (11) (2003) 4420–4427.
[188] S. Suzuki, et al., Differential roles of Epac in regulating cell death in neuronal and myocardial cells, J. Biol. Chem. 285 (31) (2010) 24248–24259.
[189] Y. Zhuang, et al., Inhibition of EPAC2 Attenuates Intracerebral Hemorrhage-Induced Secondary Brain Injury via the p38/BIM/Caspase-3 Pathway, J. Mol. Neurosci. 67 (3) (2019) 353–363.
[190] L. Zhang, et al., Inhibition of Epac2 attenuates neural cell apoptosis and improves neurological deficits in a rat model of traumatic brain injury, Front Neurosci. 12 (2018) 263.
[191] W. Liu, et al., Neuronal Epac1 mediates retinal neurodegeneration in mouse models of ocular hypertension, J. Exp. Med. 217 (4) (2020).
[192] I.M. Nijholt, et al., Neuronal AKAP150 coordinates PKA and Epac-mediated PKB/Akt phosphorylation, Cell Signal 20 (10) (2008) 1715–1724.

[193] K. Simon, et al., The orphan G protein-coupled receptor GPR17 negatively regulates oligodendrocyte differentiation via galphai/o and its downstream effector molecules, J. Biol. Chem. 291 (2) (2016) 705–718.
[194] Y.A. Syed, et al., Inhibition of phosphodiesterase-4 promotes oligodendrocyte precursor cell differentiation and enhances CNS remyelination, EMBO Mol. Med. 5 (12) (2013) 1918–1934.
[195] X. Sun, et al., Rolipram promotes remyelination possibly via MEK-ERK signal pathway in cuprizone-induced demyelination mouse, Exp. Neurol. 237 (2) (2012) 304–311.
[196] M. Schepers, et al., Amelioration of functional and histopathological consequences after spinal cord injury through phosphodiesterase 4D (PDE4D) inhibition, Neurotherapeutics (2024) e00372.
[197] M.D. Houslay, Underpinning compartmentalised cAMP signalling through targeted cAMP breakdown, Trends Biochem. Sci. 35 (2) (2010) 91–100.
[198] E. Huston, et al., EPAC and PKA allow cAMP dual control over DNA-PK nuclear translocation, Proc. Natl. Acad. Sci. 105 (35) (2008) 12791–12796.
[199] F.A. McMorris, Cyclic AMP induction of the myelin enzyme 2′,3′-cyclic nucleotide 3′-phosphohydrolase in rat oligodendrocytes, J. Neurochem 41 (2) (1983) 506–515.
[200] D.W. Raible, F.A. McMorris, Cyclic AMP regulates the rate of differentiation of oligodendrocytes without changing the lineage commitment of their progenitors, Dev. Biol. 133 (2) (1989) 437–446.
[201] C. Sato-Bigbee, G.H. DeVries, Treatment of oligodendrocytes with antisense deoxyoligonucleotide directed against CREB mRNA: effect on the cyclic AMP-dependent induction of myelin basic protein expression, J. Neurosci. Res 46 (1) (1996) 98–107.
[202] F.S. Afshari, A.K. Chu, C. Sato-Bigbee, Effect of cyclic AMP on the expression of myelin basic protein species and myelin proteolipid protein in committed oligodendrocytes: differential involvement of the transcription factor CREB, J. Neurosci. Res. 66 (1) (2001) 37–45.
[203] X. Zhang, et al., Genome-wide analysis of cAMP-response element binding protein occupancy, phosphorylation, and target gene activation in human tissues, Proc. Natl. Acad. Sci. 102 (12) (2005) 4459–4464.
[204] L. Joubert, et al., Chemical inducers and transcriptional markers of oligodendrocyte differentiation, J. Neurosci. Res. 88 (12) (2010) 2546–2557.
[205] C. Sato-Bigbee, S. Pal, A.K. Chu, Different neuroligands and signal transduction pathways stimulate CREB phosphorylation at specific developmental stages along oligodendrocyte differentiation, J. Neurochem. 72 (1) (1999) 139–147.
[206] H. Shiga, et al., Differentiation of immature oligodendrocytes is regulated by phosphorylation of cyclic AMP-response element binding protein by a protein kinase C signaling cascade, J. Neurosci. Res. 80 (6) (2005) 767–776.
[207] C. Seiwa, et al., CNS myelinogenesis in vitro: myelin basic protein deficient shiverer oligodendrocytes, J. Neurosci. Res. 69 (3) (2002) 305–317.
[208] Y. Gao, et al., Neurotrophins elevate cAMP to reach a threshold required to overcome inhibition by MAG through extracellular signal-regulated kinase-dependent inhibition of phosphodiesterase, J. Neurosci. 23 (37) (2003) 11770–11777.
[209] S.S. Grewal, R.D. York, P.J. Stork, Extracellular-signal-regulated kinase signalling in neurons, Curr. Opin. Neurobiol. 9 (5) (1999) 544–553.
[210] H.J. Yang, et al., G protein-coupled receptor 37 is a negative regulator of oligodendrocyte differentiation and myelination, Nat. Commun. 7 (2016) 10884.
[211] Z.Z. Gao, et al., EPAC negatively regulates myelination via controlling proliferation of oligodendrocyte precursor cells, Neurosci. Bull. 36 (6) (2020) 639–648.
[212] M. Malone, et al., Neuronal activity promotes myelination via a cAMP pathway, Glia 61 (6) (2013) 843–854.
[213] D.S. Gary, et al., Electrical stimulation promotes the survival of oligodendrocytes in mixed cortical cultures, J. Neurosci. Res. 90 (1) (2012) 72–83.
[214] F. De Angelis, et al., Muscarinic receptor subtypes as potential targets to modulate oligodendrocyte progenitor survival, proliferation, and differentiation, Dev. Neurobiol. 72 (5) (2012) 713–728.
[215] M. Zonouzi, et al., Bidirectional plasticity of calcium-permeable AMPA receptors in oligodendrocyte lineage cells, Nat. Neurosci. 14 (11) (2011) 1430–1438.
[216] J. Qin, et al., GD1a Overcomes Inhibition of myelination by fibronectin via activation of protein kinase a: implications for multiple sclerosis, J. Neurosci. 37 (41) (2017) 9925–9938.
[217] C. Depp, et al., Myelin dysfunction drives amyloid-beta deposition in models of Alzheimer's disease. Nature 618 (7964) (2023) 349–357.
[218] S.E. Davis, et al., The impact of neurotransmitters on the neurobiology of neurodegenerative diseases, Int. J. Mol. Sci. 24 (20) (2023).
[219] D. Paes, et al., The molecular biology of phosphodiesterase 4 enzymes as pharmacological targets: an interplay of isoforms, conformational states, and inhibitors, Pharm. Rev. 73 (3) (2021) 1016–1049.
[220] E.M. Medina-Rodriguez, et al., Inhibition of endogenous phosphodiesterase 7 promotes oligodendrocyte precursor differentiation and survival, Cell Mol. Life Sci. 70 (18) (2013) 3449–3462.
[221] R. Perez-Gonzalez, et al., Phosphodiesterase 7 inhibitor reduced cognitive impairment and pathological hallmarks in a mouse model of Alzheimer's disease, Neurobiol. Aging 34 (9) (2013) 2133–2145.
[222] Y. Chen, et al., Phosphodiesterase 7(PDE7): a unique drug target for central nervous system diseases, Neuropharmacology 196 (2021) 108694.
[223] J.X. Huang, et al., Advances in the development of phosphodiesterase 7 inhibitors, Eur. J. Med Chem. 250 (2023) 115194.
[224] E. Angelopoulou, E.S. Pyrgelis, C. Piperi, Emerging potential of the phosphodiesterase (PDE) inhibitor ibudilast for neurodegenerative diseases: an update on preclinical and clinical evidence, Molecules 27 (23) (2022).
[225] J. Prickaerts, P.R.A. Heckman, A. Blokland, Investigational phosphodiesterase inhibitors in phase I and phase II clinical trials for Alzheimer's disease, Expert Opin. Invest. Drugs 26 (9) (2017) 1033–1048.
[226] L.C. Tsai, et al., Inactivation of Pde8b enhances memory, motor performance, and protects against age-induced motor coordination decay, Genes Brain Behav. 11 (7) (2012) 837–847.
[227] C.P. Basole, et al., Treatment of experimental autoimmune encephalomyelitis with an inhibitor of phosphodiesterase-8 (PDE8), Cells 11 (4) (2022).
[228] N. Ahmad, et al., The role of Phosphodiesterase-1 and its natural product inhibitors in Alzheimer's disease: a review, Front. Pharm. 13 (2022) 1070677.
[229] L. Gomez, J.G. Breitenbucher, PDE2 inhibition: potential for the treatment of cognitive disorders, Bioorg. Med. Chem. Lett. 23 (24) (2013) 6522–6527.
[230] F.S. Menniti, T.A. Chappie, C.J. Schmidt, PDE10A inhibitors-clinical failure or window into antipsychotic drug action? Front. Neurosci. 14 (2020) 600178.
[231] P.R.A. Heckman, et al., Phosphodiesterase inhibition and modulation of corticostriatal and hippocampal circuits: Clinical overview and translational considerations, Neurosci. Biobehav Rev. 87 (2018) 233–254.
[232] M.P. Kelly, A role for phosphodiesterase 11A (PDE11A) in the formation of social memories and the stabilization of mood, Adv. Neurobiol. 17 (2017) 201–230.
[233] S.E. Anton, et al., Receptor-associated independent cAMP nanodomains mediate spatiotemporal specificity of GPCR signaling, Cell 185 (7) (2022) 1130–1142, e11.
[234] M. Zaccolo, A. Zerio, M.J. Lobo, Subcellular organization of the cAMP signaling pathway, Pharm. Rev. 73 (1) (2021) 278–309.
[235] N. Kim, S. Shin, S.W. Bae, cAMP biosensors based on genetically encoded fluorescent/luminescent proteins, Biosensors 11 (2) (2021).
[236] J.F. Zhang, S. Mehta, J. Zhang, Signaling microdomains in the spotlight: visualizing compartmentalized signaling using genetically encoded fluorescent biosensors, Annu Rev. Pharm. Toxicol. 61 (2021) 587–608.
[237] X. Zhou, S. Mehta, J. Zhang, Genetically encodable fluorescent and bioluminescent biosensors light up signaling networks, Trends Biochem. Sci. 45 (10) (2020) 889–905.
[238] D. Calebiro, I. Maiellaro, cAMP signaling microdomains and their observation by optical methods, Front Cell Neurosci. 8 (2014) 350.
[239] C. Kayser, et al., Spatiotemporal GPCR signaling illuminated by genetically encoded fluorescent biosensors, Curr. Opin. Pharm. 71 (2023) 102384.
[240] A. Bock, et al., Optical mapping of cAMP signaling at the nanometer scale, Cell 182 (6) (2020) 1519–1530, e17.
[241] A.R. Wild, M.L. Dell'Acqua, Potential for therapeutic targeting of AKAP signaling complexes in nervous system disorders, Pharm. Ther. 185 (2018) 99–121.
[242] T.B. Johnstone, et al., cAMP signaling compartmentation: adenylyl cyclases as anchors of dynamic signaling complexes, Mol. Pharm. 93 (4) (2018) 270–276.
[243] F. Klein, M.R. Machado, S. Pantano, Hitting the detection limit in cAMP signaling, Function 3 (5) (2022).
[244] J. Krzywicki, J. Koziorowska, Heliosit requires a base, Czas. Stomatol 40 (9) (1987) 606–611.
[245] S. Beca, et al., Phosphodiesterase 4D regulates baseline sarcoplasmic reticulum Ca2+ release and cardiac contractility, independently of L-type Ca2+ current, Circ. Res. 109 (9) (2011) 1024–1030.
[246] A. Robichaud, et al., Deletion of phosphodiesterase 4D in mice shortens alpha(2)-adrenoceptor-mediated anesthesia, a behavioral correlate of emesis, J. Clin. Invest 110 (7) (2002) 1045–1052.
[247] N. Amiri-Dashatan, et al., Proteomics applications in health: biomarker and drug discovery and food industry, Iran. J. Pharm. Res. 17 (4) (2018) 1523–1536.
[248] M. Cui, C. Cheng, L. Zhang, High-throughput proteomics: a methodological mini-review, Lab Invest 102 (11) (2022) 1170–1181.
[249] G. Subramaniam, et al., Integrated Proteomics Unveils Nuclear PDE3A2 as a regulator of cardiac myocyte hypertrophy, Circ. Res. 132 (7) (2023) 828–848.
[250] S. Khan, et al., Integrative omics approaches provide biological and clinical insights: examples from mitochondrial diseases, J. Clin. Invest 130 (1) (2020) 20–28.
[251] X. Wu, M.A. Hasan, J.Y. Chen, Pathway and network analysis in proteomics, J. Theor. Biol. 362 (2014) 44–52.
[252] M. Lindskog, et al., Transient calcium and dopamine increase PKA activity and DARPP-32 phosphorylation, PLoS Comput. Biol. 2 (9) (2006) e119.
[253] G. Wangorsch, et al., Time-resolved in silico modeling of fine-tuned cAMP signaling in platelets: feedback loops, titrated phosphorylations and pharmacological modulation, BMC Syst. Biol. 5 (2011) 178.
[254] F. Frohlich, et al., Mechanistic model of MAPK signaling reveals how allostery and rewiring contribute to drug resistance, Mol. Syst. Biol. 19 (2) (2023) e10988.
[255] F. Hube, et al., The phosphodiesterase inhibitor IBMX suppresses TNF-alpha expression in human adipocyte precursor cells: a possible explanation for its adipogenic effect, Horm Metab Res 31 (6) (1999) 359–362.
[256] W. Yao, et al., Rolipram, a phosphodiesterase 4 inhibitor, prevented cancellous and cortical bone loss by inhibiting endosteal bone resorption and maintaining the elevated periosteal bone formation in adult ovariectomized rats, J Musculoskelet Neuronal Interact 7 (2) (2007) 119–130.
[257] P. Christie, Roflumilast: a selective phosphodiesterase 4 inhibitor, Drugs Today (Barc) 41 (10) (2005) 667–675.
[258] Y. Zhao, H.T. Zhang, and J.M. O'Donnell, *Inhibitor binding to type 4 phosphodiesterase (PDE4) assessed using [3H]piclamilast and [3H]rolipram*, J Pharmacol Exp Ther 305 (2) (2003) 565–572.
[259] J. Schwenkgrub, et al., Ibudilast: a non-selective phosphodiesterase inhibitor in brain disorders, Postepy Hig Med Dosw (Online) 71 (0) (2017) 137–148.
[260] C.J. Tralau-Stewart, et al., GSK256066, an exceptionally high-affinity and selective inhibitor of phosphodiesterase 4 suitable for administration by

inhalation: in vitro, kinetic, and in vivo characterization, J Pharmacol Exp Ther 337 (1) (2011) 145–154.
[261] O. Bruno, et al., GEBR-7b, a novel PDE4D selective inhibitor that improves memory in rodents at non-emetic doses, Br J Pharmacol 164 (8) (2011) 2054–2063.
[262] K.B. Seamon, W. Padgett, J.W. Daly, Forskolin: unique diterpene activator of adenylate cyclase in membranes and in intact cells, Proc Natl Acad Sci U S A 78 (6) (1981) 3363–3367.
[263] Q. Cheng, Y. Zhou, Novel role of KT5720 on regulating hyperpolarization-activated cyclic nucleotide-gated channel activity and dorsal root ganglion neuron excitability, DNA Cell Biol 32 (6) (2013) 320–328.
[264] H. Hidaka, et al., Isoquinolinesulfonamides, novel and potent inhibitors of cyclic nucleotide dependent protein kinase and protein kinase C, Biochemistry 23 (21) (1984) 5036–5041.
[265] C. Liu, et al., Protein Kinase Inhibitor Peptide as a Tool to Specifically Inhibit Protein Kinase A, Front Physiol 11 (2020) 574030.
[266] C.C. Wang, et al., Calcium-Dependent Protein Kinase C Is Not Required for Post-Tetanic Potentiation at the Hippocampal CA3 to CA1 Synapse, J Neurosci 36 (24) (2016) 6393–6402.
[267] C. Sapin, L. Baricault, G. Trugnan, PKC-dependent long-term effect of PMA on protein cell surface expression in Caco-2 cells, Exp Cell Res 231 (2) (1997) 308–318.
[268] J.M. Herbert, et al., Chelerythrine is a potent and specific inhibitor of protein kinase C, Biochem Biophys Res Commun 172 (3) (1990) 993–999.
[269] H. Rehmann, Epac-inhibitors: facts and artefacts, Sci Rep 3 (2013) 3032.
[270] J.L. Best, et al., Identification of small-molecule antagonists that inhibit an activator: coactivator interaction, Proc Natl Acad Sci U S A 101 (51) (2004) 17622–17627.
[271] Y. Wang, et al., PI3K inhibitor LY294002, as opposed to wortmannin, enhances AKT phosphorylation in gemcitabine-resistant pancreatic cancer cells, Int J Oncol 50 (2) (2017) 606–612.
[272] M. Barancik, et al., SB203580, a specific inhibitor of p38-MAPK pathway, is a new reversal agent of P-glycoprotein-mediated multidrug resistance, Eur J Pharm Sci 14 (1) (2001) 29–36.
[273] D.R. Alessi, et al., PD 098059 is a specific inhibitor of the activation of mitogen-activated protein kinase kinase in vitro and in vivo, J Biol Chem 270 (46) (1995) 27489–27494.
[274] Q. Wu, et al., Selective inhibitors for JNK signalling: a potential targeted therapy in cancer, J Enzyme Inhib Med Chem 35 (1) (2020) 574–583.
[275] N.E. Uko, et al., Akt Pathway Inhibitors, Curr Top Med Chem 20 (10) (2020) 883–900.
[276] K. Ondrias, et al., [Antioxidative properties of the kampo drugs TJ-9, TJ-15, TJ-23, TJ-96 and TJ-114], Cesk Farm 41 (7-8) (1992) 253–257.