Review Article

Selective Phosphodiesterase 4D (PDE4D) Allosteric Inhibitors for the treatment of FMR-1 gene defects and other pediatric syndromal disorders associated with cognitive impairment

Abstract

The PDE4 enzymes play a crucial role in regulating signaling through the cAMP second messenger system by hydrolyzing cyclic nucleotides. The PDE4 gene family includes four subtypes, PDE4A-D, distinguished by the presence of conserved regions called UCR1 and UCR2. These enzymes can exist as dimers or monomers, with UCR1 facilitating dimerization and UCR2 controlling enzyme activity by modulating access to cAMP. Dimeric isoforms exhibit increased activity in response to cAMP signaling through PKA-mediated phosphorylation of UCR1. Mutations in the PDE4D gene have been linked to the rare neurodevelopmental disorder ACRDYS2, characterized by intellectual disability and brachydactyly. These mutations, predominantly missense mutations on the protein surface, disrupt PKA phosphorylation sites or alter interactions between UCR2 and the catalytic domain, affecting enzyme activity. Some mutations at the dimerization site increase basal enzyme activity. Genetic variations in PDE4D also influence human cognitive abilities, as evidenced by GWAS studies linking allelic variation in the gene's 5' exons encoding dimeric forms to cognitive function. This highlights the significance of dimeric PDE4D isoforms in normal brain function, both in rare disorders like ACRDYS2 and in common genetic variants associated with cognitive abilities. We focus on the role of selective phosphodiesterase 4D (PDE4D) allosteric inhibitors for the treatment of FMR-1 gene defects and other brain disorders in childhood with focus on Down syndrome, Angelman syndrome, Rett syndrome and Prader Willi syndrome.

Keywords

PDE4-inhibitor-child-brain-pediatric-syndrome

Introduction

PDE4 is an immunomodulatory target linked to cAMP. Cyclic adenosine monophosphate (cAMP) is a crucial second messenger that regulates various cellular functions, such as cell trafficking, release of inflammatory mediators, and immune cell proliferation. Drugs that increase intracellular cAMP levels inhibit immune functions of T cells, monocytes, macrophages, and neutrophils by reducing the production of pro-inflammatory cytokines and increasing the production of anti-inflammatory mediators. Intracellular cAMP levels are tightly regulated by phosphodiesterases (PDEs), a superfamily of enzymes that deactivate cAMP and cyclic guanosine monophosphate (cGMP). PDE4 is the main enzyme that degrades cAMP and consists of four subtypes (PDE4A/4B/4C/4D) with approximately 20 isoforms. The isoforms have different selectivity for cAMP and cGMP and can vary in their intracellular localization, structure, and expression in different cell types. Inhibition of PDE4 increases intracellular cAMP levels, leading to anti-inflammatory effects. The clinical

significance of different PDE4 isoforms is not fully understood, with limited information available on their functions in human tissues. Inhibition of PDE4B and PDE4D subtypes is believed to drive the anti-inflammatory effects of PDE4 inhibitors, as these subtypes are highly expressed in immune cells. Further research is needed to elucidate the roles of different PDE4 isoforms in various immune and tissue cell types. The short isoforms of PDE4B/D, particularly PDE4B2, PDE4D1, and PDE4D2, are proposed to be critical targets for achieving anti-inflammatory effects. Additional studies are required to fully understand the functional roles of different PDE4 isoforms in immune and tissue cells. The PDE4 enzymes control the spatial and temporal signaling patterns of the cAMP second messenger system by breaking down the cyclic nucleotide. The PDE4 gene family includes four subtypes, PDE4A-D, distinguished by the presence of UCR1 and UCR2 regions. PDE4 enzymes can exist as dimers or monomers, with UCR1 necessary for dimerization and UCR2 regulating enzyme activity by controlling access to cAMP. Dimeric isoforms' basal activity increases with cAMP signaling through PKA phosphorylation of UCR1. PDE4D mutations are linked to the rare neurodevelopmental disorder ACRDYS2, characterized by short fingers, intellectual disability, and reduced IQ. These mutations are surface missense mutations affecting the protein's structure and function. Genetic variations in PDE4D also influence human cognitive ability, as shown in GWAS studies associating allelic variation with cognitive function. Model organism studies demonstrate the importance of cAMP signaling in cognition and memory. PDE4 inhibitors have shown benefits in mouse models of learning, memory, depression, and neurodegeneration. Clinical use is limited by gastrointestinal side effects, but vascular inflammation seen in rodents has not been observed in humans. Current PDE4 inhibitors lack subtype selectivity and may inhibit both dimeric and monomeric isoforms of PDE4 enzymes. Recent studies focus on ameliorating cognitive impairment in different pediatric diseases. Selective phosphodiesterase 4D (PDE4D) allosteric inhibitors could play an important role in different pediatric diseases and syndromes, where impaired cognition plays a major role. The manuscript focus on this new target for pediatric patients with FMR-1 gene defects, Down syndrome, Angelman syndrome, Rett syndrome and Prader Willi syndrome.

Main Essential Data

The PDE4 enzymes are essential for regulating signaling through the cAMP second messenger system by breaking down cyclic nucleotides. The PDE4 gene family consists of four subtypes, PDE4A-D, which are distinguished by the presence of conserved regions known as UCR1 and UCR2. These enzymes can exist as dimers or monomers, with UCR1 promoting dimerization and UCR2 controlling enzyme activity by regulating access to cAMP. Dimeric isoforms show increased activity in response to cAMP signaling through PKA-mediated phosphorylation of UCR1. Mutations in the PDE4D gene have been associated with the rare neurodevelopmental disorder ACRDYS2, characterized by intellectual disability and brachydactyly. These mutations, mainly missense mutations on the protein surface, disrupt PKA phosphorylation sites or affect interactions between UCR2 and the catalytic domain, impacting enzyme activity. Some mutations at the dimerization site increase basal enzyme activity. Genetic variations in PDE4D also impact human cognitive abilities, as shown by GWAS studies linking allelic variation in the gene's 5' exons encoding dimeric forms to cognitive function. This underscores the importance of dimeric PDE4D isoforms in normal brain function, both in rare disorders like ACRDYS2 and in common genetic variants associated with cognitive abilities. Our focus is on the potential of selective allosteric inhibitors of phosphodiesterase 4D (PDE4D) for treating FMR-1 gene defects and other childhood brain disorders, particularly Down syndrome, Angelman syndrome, Rett syndrome, and Prader-Willi syndrome.

Allosteric inhibitor of phosphodiesterase 4D-BPN14770 (PDE4D)

BPN14770 is a potent allosteric inhibitor of phosphodiesterase 4D (PDE4D), with IC50 values of 7.8 nM for PDE4D7 and 7.4 nM for PDE4D3 (1,3). It has been shown to increase brain cAMP levels, enhance phosphorylation of CREB, and promote the production of brain-derived neurotrophic factor (BDNF) in the hippocampus (1,2). In a mouse model of novel object recognition (NOR), BPN14770 demonstrated cognitive benefits at doses above 0.3 mg/kg when administered orally at doses ranging from 0.1 to 30 mg/kg for 24 hours (2). The compound shows promise for the treatment of Alzheimer's disease and other brain disorders (1,2,3). Comparing wild-type and humanized PDE4D mice treated with a selective PDE4D allosteric inhibitor reveals the crucial role of PDE4D in regulating the PKA-CREB pathway. Treatment with BPN14770 increases brain cAMP levels, enhances long-term potentiation, reverses scopolamine-induced memory impairment, and improves long-term memory through a PKA-dependent mechanism. Importantly, BPN14770 is significantly more potent in enhancing memory in humanized mice compared to wild-type mice, while its effects on neurochemistry and electrophysiology show less difference (1,2,3). The memory-enhancing effects of BPN14770 persist with repeated dosing, leading to increased hippocampal CREB phosphorylation and BDNF expression (3). Markers of synaptic remodeling, such as hippocampal synapsin phosphorylation and PSD95 levels, are also elevated with repeated BPN14770 treatment (3). BPN14770 increases brain cAMP levels and enhances hippocampal long-term potentiation (LTP) in both wild-type and humanized PDE4D mice (3). Treatment with BPN14770 significantly raised cAMP levels in the brains of both wild-type and humanized PDE4D mice. LTP induction in humanized PDE4D mouse hippocampal slices was achieved with increasing concentrations of BPN14770 (1 nM, 10 nM, and 100 nM) (3).

Role of Selective Phosphodiesterase 4D (PDE4D) Allosteric Inhibitors in different pediatric syndromes

FMR-1 gene defects (FXS, FXPOI, FXTAS) in children

FMR1 disorders encompass fragile X syndrome (FXS), fragile X-associated tremor/ataxia syndrome (FXTAS), and fragile X-associated primary ovarian insufficiency (FXPOI) (2,4,12,17,20). FXS typically presents in individuals with an FMR1 full mutation or other loss-of-function variant, predominantly affecting males with developmental delay, intellectual disability, and behavioral challenges (2,4,12,17,20). Approximately 50%-70% of individuals with FXS also have autism spectrum disorder. Physical characteristics in affected males may include distinctive craniofacial features, hypotonia, gastroesophageal reflux, and other medical issues like seizures, joint laxity, and otitis media. Adults with FXS may exhibit mitral valve prolapse or aortic root dilatation. Females with the FMR1 full mutation may display similar but less frequent and milder symptoms.

FXTAS is seen in individuals with an FMR1 premutation, manifesting as late-onset cerebellar ataxia, tremors, and cognitive decline, often accompanied by psychiatric disorders (2,4,17). The onset typically occurs between 60 and 65 years, with a higher prevalence in males. FXPOI, characterized by early-onset hypergonadotropic hypogonadism, is more prevalent in women with a premutation allele compared to the general population (2,4,12,17,20). Specialized molecular genetic testing is essential for diagnosing FMR1 disorders, focusing on detecting CGG trinucleotide repeat expansions in the FMR1 gene. A full-mutation repeat size (>200 CGG repeats) is indicative of FXS, while a premutation-sized repeat (55-200 CGG repeats) is associated with FXTAS or FXPOI. Comprehensive genomic testing is recommended when FXS is suspected but no CGG repeat

expansion is detected. Treatment strategies for FMR1 disorders involve supportive care tailored to individual needs, including psychopharmacologic interventions, behavioral therapies, speech and occupational therapy, and educational support for FXS. Symptomatic and personalized approaches are recommended for FXTAS, while gynecologic or reproductive endocrinologic evaluations are crucial for managing FXPOI and addressing reproductive and hormonal concerns. FMR1 disorders should be considered in individuals presenting with the following clinical and associated findings. Fragile X syndrome (FXS) is found in males and females with unexplained intellectual disability or developmental delay; in males with unexplained autism spectrum disorder and females with unexplained autism spectrum disorder, along with additional indicators such as a phenotype compatible with FXS, family history of X-linked neurodevelopmental disorders, or premature ovarian failure, ataxia, or tremors in close relatives. Fragile X-associated tremor/ataxia syndrome (FXTAS) is found in males and females experiencing late-onset intention tremor and cerebellar ataxia of unknown cause; in men and women with dementia may also be considered if ataxia, parkinsonism, or tremor are present; and males and females with multiple system atrophy, cerebellar subtype, especially with a prolonged course. Fragile X-associated primary ovarian insufficiency (FXPOI) was found in females with unexplained primary ovarian insufficiency or failure before age 40 years. FMR1 disorders are diagnosed through specialized molecular genetic testing with signs of full-mutation repeat size (>200 CGG repeats) is typically required for a definite diagnosis of FXS and a remutation-sized repeat (55-200 CGG repeats) is associated with FXTAS or FXPOI. The allele size in normal alleles are approximately 5-44 repeats, with little instability. Intermediate alleles are approximately 45-54 repeats, may expand into the premutation range. Premutation alleles are pproximately 55-200 repeats, associated with increased risk for FXTAS and FXPOI. Full-mutation alleles consist of more than 200 CGG repeats, typically associated with aberrant hypermethylation of the FMR1 promoter and somatic variation in repeat number. Selective Phosphodiesterase 4D allosteric inhibitors could have a potential in these rare pediatric disease group of FMR-1 gene defects. Further evaluation is necessary to get more information about the cognitive status of these patients with this target.

Down Syndrome

Trisomy 21 or Down syndrome is a chromosomal anomaly that manifests as a combination of intellectual disability and physical abnormalities of varying degrees. The cause lies in a peculiarity in the genetic makeup of the affected individual (genomic mutation, chromosomal aberration, or aneuploidy). In this case, chromosome 21 (chromosomes are components of cells where genetic information is stored) or parts of it are present in triplicate instead of duplicate. This chromosomal disorder is therefore called Trisomy 21 (from the ancient Greek words τρία tría, meaning 'three, threefold' and σῶμα sôma, meaning 'body'; here: chromosome body No. 21, as the carrier of genetic information). Triplications of chromosomes can occur when cell division results in two chromosomes of the same number entering the germ cell instead of one. The fertilized egg cell then has a total of three chromosomes: one from the mother, one from the father, and an additional one from either the mother or the father. The likelihood of a child having Trisomy 21 increases with the mother's age

(especially from the age of 35 onwards). Individuals with Down syndrome typically exhibit characteristic physical features and are usually impaired in their cognitive abilities, meaning their thinking abilities. The abnormalities and impairments can vary in severity. The triplication of the corresponding genetic material usually occurs through missegregation, the failure of chromosomes to separate during meiosis (cell division). The different forms of Trisomy 21 can either occur spontaneously or be inherited if the mother herself already has Down syndrome. However, a translocation trisomy (due to chromosomal rearrangement) can occur more frequently in families if one parent is predisposed, meaning a balanced translocation of a 21st chromosome is present in a parent without the symptoms of Down syndrome. A definitive prenatal diagnosis is currently possible through an examination of fetal chromosomes obtained through amniocentesis (amniotic fluid sampling) or chorionic villus sampling (placental sampling), less commonly through umbilical cord sampling (cordocentesis). Since 2012, traces of fetal genetic material can be found and examined through blood tests in the mother. The potential selection through pregnancy termination is ethically controversial. In Germany, there are approximately 30,000 to 50,000 people living with Trisomy 21. Trisomy 21 is the most common chromosomal aberration in newborns. Statistical surveys show that the probability of having a child with Trisomy 21 exponentially increases with the mother's age: at the age of 25, it is less than 0.1 percent, at 35 it is 0.3 percent, at 40 it is one percent, and at 48 it is nine percent. This is likely due to the aging of the woman's prenatal eggs, which leads to more errors in meiosis. Depending on the maternal age distribution in a population, the average prevalence is between 0.125 to 0.2 percent, or about 1 in 500 to 1 in 800. In Germany, the expected proportion of children with Down syndrome in 2006 was about 1 in 500, or 0.2 percent. The increasing prevalence of Trisomies in children with the mother's age does not correspond proportionally to the number of children actually born with Trisomy 21 in the Western hemisphere. This is because older mothers are more likely to undergo prenatal diagnosis. If a high probability of Trisomy 21 is detected, mothers in most cases choose to terminate the pregnancy. A statistical survey between 1992 to 1996 showed that about a quarter of children with Trisomy 21 in the 25- to 29-year-old age group and about a third in the 30- to 34-year-old age group were already detected prenatally. After diagnosis and counseling, 94.5 percent of mothers did not carry their child to term. Another study from 1988 to 1997 reported an overall prenatal detection rate of 53 percent. In the total group, only 23 percent of mothers were 35 years or older (with 77 percent of them not expected to have an increased likelihood of Trisomy 21 in their child). After prenatal diagnosis, 90 percent of mothers did not carry their child to term. In the summer of 2012, a new non-invasive prenatal test for Down syndrome was introduced. On the other hand, the pressure to undergo prenatal diagnosis with the option of abortion increases. It is becoming increasingly difficult to wholeheartedly embrace one's unborn child for personal, even religious reasons." In many European countries, very few children with Down syndrome are born due to the high rate of abortions. Boys are more affected than girls (androtropy). Dittmann found a ratio of 53:47, Wilken observed ratios of 57.2:42.9 (1974) and 54:46 (2000). The reason for this imbalance is not yet clear. In addition to a familial predisposition for multiple pregnancies, the likelihood of having twins or higher-order multiples increases with the mother's age. Hormone treatment and assisted reproductive techniques to increase fertility also increase the likelihood of multiples. The combination of these developments with the natural increase in the likelihood of Down syndrome in older mothers means that the rate of births of children with Trisomy 21 in multiple pregnancies also increases. In the National Down Syndrome Cytogenetic Register in the UK, there were a total of 244 pairs of twins registered in 2003; 29 pairs of them (11.8 percent) had both children with Trisomy 21 (one pair was fraternal: girl/boy). Additionally, nine sets of triplets were known, each with one child having an extra chromosome 21. A definitive diagnosis is currently only possible through an examination of the chromosomes themselves, traditionally obtained through amniocentesis or chorionic villus sampling, less commonly through umbilical cord puncture (cordocentesis). These are invasive procedures, each

associated with a different risk of procedure-related miscarriages. Since 2012, novel non-invasive test procedures have been approved in many countries, which analyze the mother's blood for traces of fetal genetic material and now respond to trisomies 21, 18, and 13. Researchers at Stanford University in California claimed in 2008 to have successfully amplified cell-free DNA fragments of the mother and fetus from maternal blood and assigned them quantitatively to the 46 human chromosomes. If the blood samples come from pregnant women whose fetuses have a trisomy, the DNA of their additional chromosomes is more strongly represented in the maternal blood than in unaffected pregnancies. This higher presence could be demonstrated by the novel method for various chromosomal abnormalities, including Trisomy 21. Early on, it was discussed whether such non-invasive methods of prenatal diagnosis would replace or at least significantly reduce the frequency of invasive methods in a few years. By the end of 2014, a provider reported approximately 10,000 tests conducted by their company, about half of which were on German women. In May 2015, a Swiss newspaper reported about 4,500 tests on Swiss women and the first-time coverage of costs by a health insurance company. The non-invasive procedures have a very low error rate according to the manufacturers; however, prenatal diagnosticians recommend confirming a positive diagnosis through one of the traditional invasive methods. The approval of non-invasive procedures has been and continues to be strongly criticized on ethical grounds. Disability advocates call for a ban on the tests, as they lower the threshold for such examinations and subsequent abortion of affected fetuses and ultimately violate the right to life of disabled individuals; furthermore, they pave the way for further fetal genetic testing and "designer babies." Supporters, on the other hand, point to approximately 98% of normal test results - in these cases, the test avoids the risks associated with invasive methods for the fetus and mother. Recommendations from manufacturers to use the tests for prenatal trisomy screening in pregnant women of all ages have fueled the ethical debate, including whether the tests should be funded by statutory health insurance. In Down syndrome, amyloid precursor protein levels induce Alzheimer similar symptoms. Selective phosphodiesterase 4 D allosteric inhibitors could play a role in Down syndrome patients, who develop in later life a form of dementia or Alzheimer disease to ameliorate the cognitive impairment in these elder Down patients (23).

Angelman Syndrome

The Angelman syndrome is the result of a rare genetic change on chromosome 15 (microdeletion on the maternal chromosome or uniparental disomy 15q11-13. It is often associated with developmental delays, cognitive impairment, above-average happiness, and severely reduced speech development. The British pediatrician Harry Angelman first described the syndrome under scientific terms in 1965, naming it the Happy Puppet Syndrome due to the distinctive movement patterns and frequent laughter of the children he cared for. The life expectancy of individuals with Angelman syndrome is not reduced. Both boys and girls can be affected by Angelman syndrome. In 1965, Angelman described 150 case studies; by 2005, over 800 cases were known worldwide. The syndrome occurs with an average frequency of 1 in 15,000 to 1 in 20,000. Over time, various characteristics have been documented that are frequently present in individuals with Angelman syndrome. Not

all affected individuals exhibit all characteristics, and the existing features do not occur in all individuals to the same extent: Frequent, often objectively unfounded smiling and laughter (unmotivated laughter), sometimes actual laughing fits, often during excitement and stress. Hyperactivity and cognitive impairment is often found. Difficulty concentrating, short attention span, but often good memory for faces and directions and good spatial orientation are typical features of Angelman children. In early childhood, often no attempts at speech, no babbling, later only very limited ability for expressive speech articulation, but some ability to learn alternative forms of communication, e.g., sign language using the system of augmentative and alternative communication (AAC) and picture communication will be peformed. The have a good receptive language (language comprehension). They have movement and balance disorders, ataxia (usually a rather stiff, clumsy, swaying, wide-legged gait, jerky, choppy (walking) movements. One in ten children does not learn to walk. Children with Angelman syndrome have delayed motor development (resulting in relatively late walking) and sensory disturbances in the physical area (often, for example, balance problems). In Angelman syndrome, children develop cognitive impairment with memory delay. Selective phosphodiesterase 4 D allosteric inhibitors could play a role in Angelman syndrome patients to ameliorate cognitive functions of the brain. To date, there are no clinical studies found in this aspect.

Rett Syndrome

Rett syndrome is a profound developmental disorder (24-32). The cause is an encephalopathy that follows an X-chromosomal dominant inheritance pattern (24-32). X-chromosomal mutations occur at the time of conception in both male and female embryos. However, in male embryos, the mutations almost always lead to intrauterine fetal death due to hemizygosity. Therefore, almost exclusively girls with Rett syndrome are observed (gynatropia). Rett syndrome was first described in 1966 by Viennese pediatrician Andreas Rett (1924–1997) (24-32). In Germany, the frequency is estimated to be 1:15,000 to 1:10,000. Affected children initially develop seemingly normally (25,27,28). Between the seventh month and the second year of life, the child loses, after a variable phase of developmental stagnation, at least partially already acquired skills, especially speech and hand use. The condition of the children then stabilizes again, and reaching a normal age is possible. People with Rett syndrome typically exhibit symptoms of autism and movement coordination disorders (ataxia). Some affected individuals have intellectual disabilities, many speak a few words and can follow simple commands. Epileptic seizures and hand stereotypies, which resemble handwashing movements, are also characteristic of Rett syndrome (24-32). In 1998, the cause of Rett syndrome was localized at Baylor College of Medicine in Houston and Stanford University (24,27,30). The cause is a mutation of the MECP2 gene (24-32). Since October 1999, Rett syndrome can be diagnosed using a genetic test that can be administered very early in the child's development. In about 80 to 90% of cases, it involves dominant de novo mutations of the X chromosome (germline mutation), which mainly occur in the male germline and are therefore mainly passed from father to daughter (sons receive the Y chromosome from the father). The MeCP2 gene (Methyl-CpG-Binding Protein 2) is located in the chromosomal region Xq28. MeCP2 is a transcription factor that selectively binds to methylated CpG islands and represses the transcription of various genes. This disrupts cholesterol metabolism significantly (24-32). Rett discovered the typical hand movements (washing movements) in 1965 when two young girls were sitting on their mothers' laps in the waiting room of his practice, and their mothers accidentally released their daughters' hands at the same time. These hand stereotypies are now considered the most typical criterion for Rett syndrome. Over time, additional diagnostic criteria were added. Working on CRISP-Cas 9 technology interesting studies were published curing Rett syndrome patients by gene editing

method (27). Selective phosphodiesterase 4 D allosteric inhibitors could play a role in Rett syndrome patients to ameliorate cognitive functions of the brain they lost over time. Studies on Rett patients to treat them with capsules of selective PDE4D allosteric inhibitors should be initiated.

Prader Willi syndrome

Prader-Willi syndrome (PWS), also known by the synonyms Prader-Labhard-Willi-Fanconi syndrome, Urban syndrome, and Urban-Rogers-Meyer syndrome, is a relatively rare disability caused by a damaged human chromosome 15 (36-40). It is based on an inherited gene mutation or a mutation-related error in the genomic imprinting mechanism of chromosome 15 (microdeletion syndrome) and is associated with physical, metabolic, and cognitive symptoms caused by dysfunction of the hypothalamus (36-40). Babies with Prader-Willi syndrome have reduced movement, muscle weakness, low birth weight, and slow weight gain immediately after birth (36,38,40). They do not start crying or cry very weakly and often have sucking and swallowing difficulties. The original description of the symptoms dates back to John Langdon Down, who also described children with Williams-Beuren syndrome and became known for the detailed description of Down syndrome named after him. A 21-year-old woman with Prader-Willi syndrome was described by him in 1864 (36-40). Prader-Willi syndrome was first detailed under scientific aspects in 1956 by the Zurich pediatricians Andrea Prader, Alexis Labhard, and Heinrich Willi (36-40). They explained the individual symptoms but could not make statements about the causes. In 1981, it was found that Prader-Willi syndrome is caused in about 70% of cases by an incomplete paternal chromosome 15. Prader-Willi syndrome usually occurs sporadically and affects approximately one in 10,000 to 15,000 children. Girls and boys are equally affected. In some cases, familial clustering and sibling cases have been described. The cause of the syndrome is that the gene copy inherited from the father is incomplete or non-functional. The chromosomal region 15q11-13 undergoes genomic imprinting, meaning that certain genes on this segment are active only on the chromosome inherited from the father and others are only active on the one from the mother. In Prader-Willi syndrome, certain paternal genes are not expressed, and the corresponding ones on the maternal chromosome are silenced, resulting in a complete lack of the gene product. In recent years, various causes at the chromosomal level have been identified through more precise genetic marking techniques. These include the aforementioned paternal deletion, where a piece of the paternal chromosome 15 is missing (microdeletion), maternal disomy, where two maternal chromosome 15s are present and the father's is missing (uniparental disomy 15), or an error in the genomic imprinting of the chromosome segment. Prader-Willi syndrome is one of the conditions where a functional loss of genes subject to genomic imprinting is typically identified as the cause. If the deletion affects the chromosome 15 segment inherited from the mother rather than the father, it leads to Angelman syndrome. This genetic peculiarity is more common in the familial environment of people with Prader-Willi syndrome. Various genes that have been lost through deletion, such as the SNRPN gene and the Necdin gene, are discussed as triggers for Prader-Willi syndrome. The observable effects of Prader-Willi syndrome can largely be attributed to a lack of hormone release (gonadotropin-releasing hormone) in the hypothalamus caused by the described genetic peculiarities. The faulty hormone release affects other hormone-producing glands, such as the thyroid, adrenal glands, and gonads (testes and ovaries). A study suggested that Prader-Willi syndrome patients do not express a specific snoRNA (HBII-52), leading to disrupted splicing regulation of the serotonin receptor 5-HT2CR. This could be a possible reason for the patients' good response to selective serotonin reuptake inhibitors (SSRIs). However, another study shows that HBII-52 does not play a significant role in Prader-Willi syndrome. The absence of SNORD116 (formerly HBII-85) snoRNAs leads to the main symptoms of

Prader-Willi syndrome and is considered the main cause. Selective phosphodiesterase 4 D allosteric inhibitors could play a role in Prader Willi syndrome patients to ameliorate cognitive functions of the brain. To date, there are no clinical studies found in this aspect. Further clinical investigations should focus on studying the cognitive function after orally administered treatment with PDE4D allosteric inhibitors in these group of patients.

Discussion

The enzymes of the cyclic nucleotide phosphodiesterase family (PDE) catalyze the hydrolytic cleavage of the second messengers cAMP and cGMP (cyclic adenosine and guanosine monophosphate). They are part of a control system that regulates the activity of these important signaling molecules and lead to the shutdown of induced signaling cascades. The PDE enzymes thus act as counterparts to the adenylate and guanylate cyclases, which synthesize the cyclic nucleotides (after activation). cAMP and cGMP regulate a variety of effector proteins, including protein kinase A and G, cyclic nucleotide-gated ion channels, transcription factors, and PDE enzymes that have allosteric binding sites in addition to the catalytic ones. The cAMP and cGMP signaling systems are among the first discovered signal transduction pathways and control a variety of physiological and pathophysiological processes. These include gene expression, cell proliferation and differentiation, apoptosis, inflammation, carbohydrate and lipid metabolism. A large number of drugs target this network of signaling pathways with the cAMP/cGMP junction. For example, while agonists at Gαs-coupled receptors directly stimulate the activity of adenylate cyclase and thus the synthesis of cAMP, the effect of caffeine is partly due to inhibition of PDE, which also leads to a higher intracellular level of cyclic nucleotides. Other methylxanthines such as theophylline have historically been used in the treatment of asthma, even before the mechanism of action and the influence on PDE were understood. It is now known that this class of compounds acts as non-selective PDE inhibitors. Due to an unfavorable risk-benefit profile (and better alternatives), they are now only used to a limited extent. In the 1970s to 1980s, scientists were able to characterize the first PDE isoforms using more advanced biochemical methods with known compounds (with varying selectivity). Over time, a picture emerged of eleven PDE families and their activation, affinity, and selectivity towards cAMP, cGMP, and synthetic modulators. These eleven PDE families are structurally related but have functional differences. Through the interplay of different genes, alternative splicing, and transcriptional processing, nearly 100 isoforms can be formed. The N-terminal part of the PDE molecules determines their association with the respective compartments and signalosomes and is therefore highly variable. Signalosomes are macromolecular complexes based on protein-protein interactions that regulate the signal transduction of cyclic nucleotides. The catalytic center, on the other hand, is largely conserved and has a similar structure of 16 helices in all crystallized PDE families. These form a hydrophobic pocket and the catalytic histidine-containing motif that includes binding sites for two divalent metal ions. PDE inhibitor co-crystals suggest that a highly conserved glutamine (hydrogen bonding, orientation of inhibitors) and phenylalanine are essential for inhibitor binding. To develop subtype-selective compounds, researchers exploit differences in the rigidity of the binding pocket and minimal size differences. A specific situation exists with PDE5: Due to the flexibility of two loop motifs, the protein can adopt different conformations. This explains the 5- to 20-fold difference in binding affinity of sildenafil and vardenafil, as they bind in different conformations. Highly selective PDE inhibitors are increasingly being developed through structure-based design and using high-throughput crystallography. New approaches in the development of PDE inhibitors and their signalosomes focus mainly on protein-protein interactions between the PDE isoenzymes and their regulatory

partners. Preclinically, it has been shown that different subtypes of PDE4 are strongly expressed in cells involved in immuno-inflammatory processes. Consequently, PDE4 inhibitors have demonstrated potent anti-inflammatory effects in numerous cell and animal models. Based on these findings, a potential use in inflammation-driven lung diseases has emerged, reflected in the approval of Roflumilast in 2010 (EMA) and 2011 (FDA). In recent years, this first approved PDE4 inhibitor has increasingly been used in patients with chronic obstructive pulmonary disease (COPD). Roflumilast is used as an oral therapy in combination with a bronchodilator for severe COPD with concomitant chronic bronchitis and frequent exacerbations. A new PDE4 inhibitor, Apremilast (Otezla®), has recently gained attention due to positive study data (33,34,35). In the Phase III study PALACE-I, this substance showed improvement in Psoriatic Arthritis (ACR20 response) in 31% (20 mg Apremilast, twice daily) and 40% (30 mg Apremilast, twice daily) of patients compared to 19% in the placebo group (33). Additionally, 34% and 51% of patients (compared to 19% under placebo) experienced a reduction of psoriasis-affected skin area and severity of lesions by more than 50% (PASI-50 response). This study and data from ongoing studies PALACE-II and -III showed a mild side effect profile with diarrhea, nausea, and headaches that were dose-dependent. Apremilast was approved by the FDA in March 2014 for patients with Psoriatic Arthritis (33-35). Recent studies showed, that PDE4D inhibitors like BPN14770 seems to have positive effects for cognition in pediatric impaired children, and therefore should be further evaluated in the pediatric subgroups of syndromes like Down-, Angelman-, Rett- or Prader Willi syndrome.

Conclusion

Extensive experimental and clinical research indicates that targeting PDEs with pharmacological agents could be a promising approach for treating cognitive impairments. PDE inhibitors regulate levels of cyclic purine nucleotides, potentially offering preventive or therapeutic benefits for Alzheimer's disease in adults, mild cognitive impairment, and dementia in adults. Dysregulation of various cellular processes, including immune response, signaling pathways, and inflammation, may be linked to abnormal PDE activity, which is implicated in neurodegenerative disorders. This review discusses different types of PDE4D inhibitors. The protective effects of these medications, attributed to PDE inhibition and other pharmacological actions like neurotransmitter receptor modulation or vasodilation, are highlighted. PDEs are emerging as promising targets for developing new drugs to treat cognitive impairment in rare syndromes in childhood mentioned above, but well-designed clinical trials are necessary to evaluate their efficacy and safety in this small pediatric population.

Ethical Approval: The author has adhered to the accepted ethical standards.

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