Molecules and Cells

Minireview

Proline Metabolism in Neurological and Psychiatric Disorders

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Proline plays a multifaceted role in protein synthesis, redox balance, cell fate regulation, brain development, and other cellular and physiological processes. Here, we focus our review on proline metabolism in neurons, highlighting the role of dysregulated proline metabolism in neuronal dysfunction and consequently neurological and psychiatric disorders. We will discuss the association between genetic and protein function of enzymes in the proline pathway and the development of neurological and psychiatric disorders. We will conclude with a potential mechanism of proline metabolism in neuronal function and mental health.

Keywords: cell metabolism, neurological disease, neuron, proline, psychiatric disease

INTRODUCTION

The prevalence of neurological and psychiatric disorders was approximately 1.28 billion as of 2017, accounting for 17.4% of the global disease burden (GBD 2017 Disease and Injury Incidence and Prevalence Collaborators, 2018). Psychiatric disorders frequently accompany neurological disorders, e.g., Alzheimer's disease (AD), Parkinson's disease (PD), epilepsy, migraine, essential tremors, and stroke (Kupeli et al., 2021). Both neurological and psychiatric disorders are associated with impaired neurotransmission, altered brain structure, inflammation, and impaired neurotrophic signaling (Kupeli et al., 2021). Treatments targeting these abnormalities appear to be effective but with significant adverse side effects, and more efficacious therapies for neurological and psychiatric disorders remain an unmet need.

In this review, we focus on the nonessential amino acid proline, the accumulation of which leads to hyperprolinemia. As early as 1989, a hyperprolinemia patient's brain showed a low density of white matter (Steinlin et al., 1989), which indicates a potential role of proline metabolism in neuronal morphology and function.

NEURONAL METABOLISM

Glucose is the primary substrate of neuronal metabolism. Neuronal cells take up glucose through specific glucose transporters, which are then phosphorylated by hexokinase to produce glucose-6-phosphate (Dienel, 2019). Glucose-6-phosphate goes through different metabolic pathways and eventually becomes oxidized to CO_2 and H_2O in the brain (Dienel, 2019). However, as glucose can follow different metabolic routes, each neuron does not necessarily metabolize glucose to produce CO_2 and H_2O (Zielke et al., 2009). Indeed, glucose can form a wide range of metabolic intermediates, such as lactate, pyruvate, glutamate, and acetate, in neurons. These intermediates can subsequently be oxidized for energy production, serve as neurotransmitters and so on (Zielke et al., 2009). Damage to these processes can lead to neurological disturbances, loss of consciousness, and even coma within

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minutes (Belanger et al., 2011).

Lipids comprise approximately 50% of the brain's dry weight. They are the key components that support the brain's complex structure and function (Barber and Raben, 2019). It has long been known that lipid component fatty acids can enter the brain and be oxidized (Allweis et al., 1966; Dhopeshwarkar and Mead, 1970). There is evidence that approximately 20% of the total energy expenditure of the adult brain is provided by the oxidation of fatty acids (Ebert et al., 2003). Moreover, long-chain polyunsaturated fatty acids (PUFAs) have been proven to be the precursors of several metabolites with different effects on inflammation and neuron outgrowth (Martinat et al., 2021). Omega-3, a PUFA, is a potential treatment for schizophrenia (Frajerman et al., 2021). Except for the fatty acid itself, a few lipid metabolism enzymes have been identified at synaptic terminals, where they can locally modulate synaptic transmission (Di Paolo et al., 2004; Rohrbough et al., 2004). Unlike adipocytes and liver cells, neurons are not energy-storage cells. Thus, they do not normally contain significant amounts of nonstructural lipids and receive metabolites oxidized from fatty acids, such as ketones, NADH, acetyl-CoA, and $FADH_2$, from astrocytes (Barber and Raben, 2019).

Amino acids are better known for their roles in producing neurotransmitters than providing energy. Glutamate is the most abundant amino acid in the mammalian brain and functions as an excitatory neurotransmitter (Clark and Amara, 1993). Aspartate was later identified as an excitatory neurotransmitter (D'Aniello et al., 2011). γ-Aminobutyric acid (GABA) and glycine are inhibitory neurotransmitters (Clark and Amara, 1993). Except for these amino acids that serve as neurotransmitters, the functions of the other amino acids, such as proline, are less well understood. It is interesting to note that accumulating evidence now indicates that proline is associated with neurological and psychiatric disorders (Mayneris-Perxachs et al., 2022; Mitsubuchi et al., 2014).

Most nutrient metabolism occurs in the mitochondria, the powerhouse of the cell. Mitochondria are highly dynamic organelles that divide, fuse, and move purposefully within axons and dendrites (Mattson et al., 2008). In neurons, mitochondria are the major sites for energy production, generation of reactive oxygen species (ROS), calcium signaling, developmental and synaptic plasticity, and the arbitration of cell survival and death (Srivastava et al., 2018). Many gene products are localized in the mitochondria, and mutations of these genes have been linked to neurological and psychiatric diseases (Franco et al., 2021; Iwamoto et al., 2005; Sreedharan et al., 2008). Mitochondria-mediated oxidative stress perturbs Ca^{2+} homeostasis, and apoptosis also contributes to the pathogenesis of prominent neurological diseases, including AD, PD, Huntington's disease, stroke, amyotrophic lateral sclerosis (ALS), and psychiatric disorders (Chan, 2020; Falabella et al., 2021). Overall, nutrient metabolism in mitochondria has a profound effect on neuronal functions, and prolonged defects eventually lead to neurological and psychiatric disorders (Fig. 1).

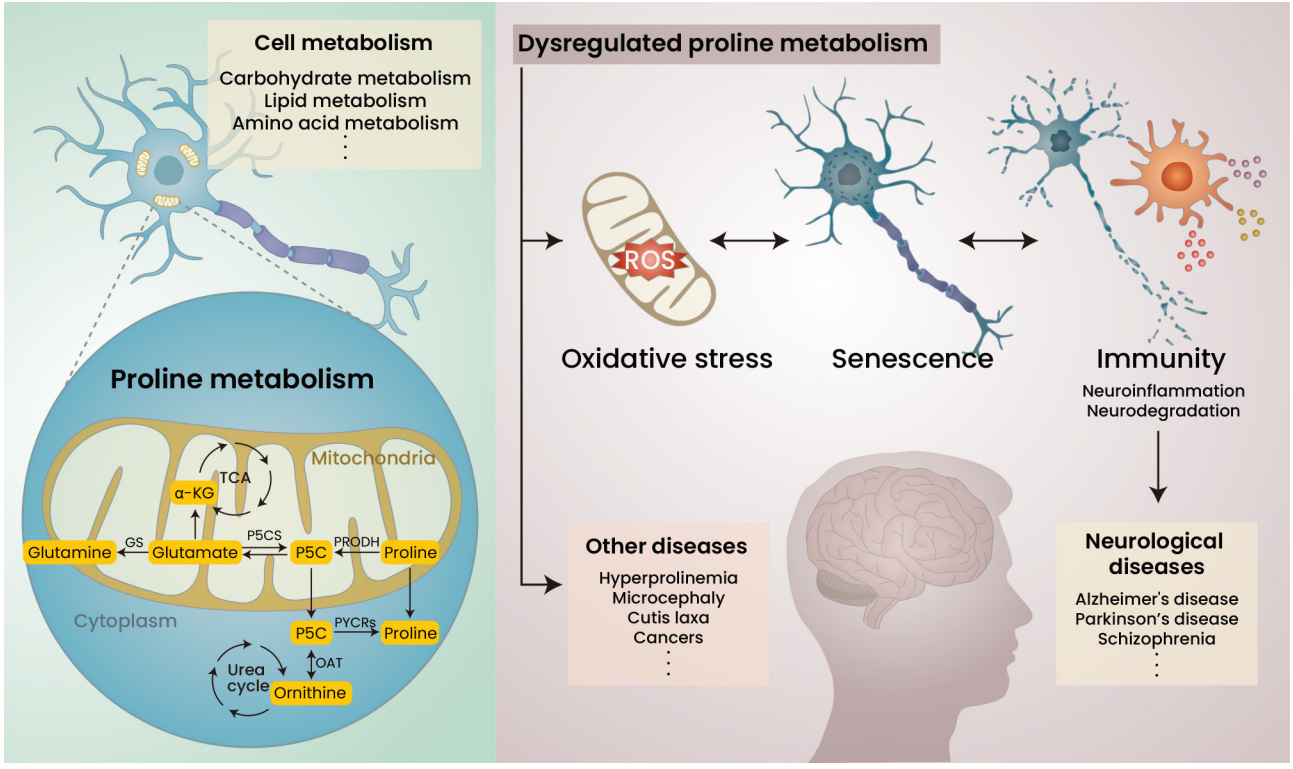

Fig. 1. Overview of proline metabolism in neurological and psychiatric disorders.

NEURONAL DYSFUNCTION IN NEUROLOGICAL AND PSYCHIATRIC DISORDERS

AD is the most common neurodegenerative disorder and the leading cause of dementia in the elderly (Palop and Mucke, 2010). The hallmark of AD is the accumulation of pathogenic amyloid-β (Aβ) assemblies in the brain, resulting in the progressive dismantling of synapses, neuronal circuits, and networks (Querfurth and LaFerla, 2010). In AD patients, hippocampal synapses begin to decline, and synaptic profiles show compensatory increases in size. They also show a reduction of approximately 25% in the presynaptic vesicle protein synaptophysin (Querfurth and LaFerla, 2010). Moreover, abnormal axons associated with neurofibrillary tangles and abnormal thin and tortuous axons were shown in patients' brains (Stokin and Goldstein, 2006). *In vitro* studies showed that elevated Aβ attenuates excitatory synaptic transmission by decreasing the number of surface AMPA receptors and NMDA receptors, which is associated with a collapse of glutamatergic dendritic spines (Hsieh et al., 2006; Shankar et al., 2007).

PD is a progressive neurodegenerative disease that affects 2%-3% of the population ≥65 years of age (Poewe et al., 2017). The main neuropathological hallmarks of PD are dopaminergic neuronal loss in the pars compacta of the substantia nigra (SNc) and accumulation of intraneuronal inclusions called Lewy bodies and Lewy neurites (Calabresi et al., 2013). However, it is not clear what causes the death of dopaminergic neurons. One hypothesis is that accumulated mitochondrial DNA damage produced by ROS and related free radicals generated in oxidative phosphorylation would lead to the dysfunction of Ca^{2+} channels for pacemaking. SNc dopamine neuron activity relies on Ca^{2+} channels. This reliance may cause persistent metabolic stress on mitochondria, accelerating SNc dopamine neuron aging and death (Surmeier, 2007). In addition to the Ca^{2+} channel and mitochondrial hypothesis, new insights have suggested that inflammation causes brain blood barrier dysfunctions and gut-brain connections (Bartels et al., 2020; Sweeney et al., 2019; Travagli et al., 2020).

Schizophrenia is a severe psychiatric disorder that has a profound impact on the individual and society (Owen et al., 2016). Diagnosis is made clinically based on history and mental status examination. There are no diagnostic tests or biomarkers (Owen et al., 2016). However, one of the most consistent findings in schizophrenia is ventricular enlargement and decreased brain volume involving the hippocampus and cortex (Bellon, 2007). There are many hypotheses for schizophrenia. The first is the dopamine hypothesis. Several clinical studies reported that schizophrenia patients showed elevated presynaptic striatal dopamine synthesis, while other studies reported either a small but not significant elevation or a small reduction in dopamine levels (Howes and Kapur, 2009). On the other hand, dopamine receptors also showed abnormal expression; however, studies were inconsistent (Howes and Kapur, 2009). The second is the glutamate hypothesis. In addition to NMDAR antagonist studies, brain imaging studies, and genetic studies, postmortem studies are the most direct evidence. In patients, postmortem studies revealed a significant reduction in GABA levels in the prefrontal cortex, a reduced number of GAD67-expressing neurons that coexpress NR2A in the prefrontal cortex, and reduced activity of glutamate carboxypeptidase II in the frontal cortex, hippocampus, and temporal cortex (Coyle, 2006; Uno and Coyle, 2019). Other hypotheses, such as serotonin, inflammation and two-hits, have also been proposed (Eggers, 2013; Feigenson et al., 2014). More evidence and clear hypotheses are needed for the diagnosis and treatment of schizophrenia.

PROLINE METABOLISM IN NEUROLOGICAL AND PSYCHIATRIC DISORDERS

Proline is a nonessential amino acid involved in adaptation to osmotic and dehydration stresses, redox control, and apoptosis (Fichman et al., 2015). Proline is catabolized to pyrroline-5-carboxylate (P5C) by proline dehydrogenase (PRODH) in the mitochondrial matrix, generating glutamate through NAD-dependent P5C dehydrogenase (P5CDH). Glutamate is an important excitatory neurotransmitter in neurons and a precursor to glutamine, GABA and mitochondrial TCA cycle intermediates. Conversely, glutamate is converted into a P5C intermediate through P5C synthase (P5CS) and further reduced to proline by P5C reductases (PYCRs). All of the enzymes in the proline metabolism pathway have been reported to be associated with neurological or psychiatric disorders in human and animal models (Table 1) (Guo et al., 2018; Li et al., 2021; Liu et al., 2009).

A high-throughput metabolomics study reported that proline metabolism plays a key role in the progression from healthy to mild cognitive impairment to AD in a natural aging population (Xie et al., 2021). The disease-metabolite-pathway associations highlight the importance of proline in all three neurodegenerative diseases, including AD, PD, and ALS (Kori et al., 2016). Clinically, patients with P5CS deficiency present profound manifestations with progressive neurodegeneration, mental retardation, peripheral neuropathy, joint laxity and cataracts (Baumgartner et al., 2000; Phang et al., 2013). Twenty-four hypomyelinating leukodystrophy patients with mutations in the proline synthase gene, PYCR2, displayed postnatal progressive microcephaly, poor overall growth, and severely delayed psychomotor development. Neuroimaging analysis of these patients revealed a decreased myelin sheath, thinning of the corpus callosum and brainstem, and a general decrease in white matter volume (Escande-Beillard et al., 2020). The proline-degrading enzyme PRODH is located on chromosome 22q11, a schizophrenia locus. At least eight single nucleotide polymorphisms have been reported in the PRODH gene in patients with schizophrenia (Bender et al., 2005; de Koning et al., 2015; Ota et al., 2014), and one hyperprolinemia patient was found to have white matter abnormalities (Steinlin et al., 1989). Interestingly, hydroxyproline and proline concentrations in urine are influenced by stress and anxiety (Lee et al., 2011).

In animal models, *Pycr2* knockout mice developed progressive neurological symptoms, including white matter defects, marked developmental delay, and premature death (Escande-Beillard et al., 2020). Glutamate, GABA and aspartate were significantly reduced in the hypothalamus and frontal

Table 1. Proline metabolism in diseases

Gene/enzyme names	Type of mutations	Disease/phenotype	References
P5CS	Deletion	Progressive neurodegeneration	(Baumgartner et al., 2000)
		Mental retardation	(Baumgartner et al., 2000)
		Peripheral neuropathy	(Baumgartner et al., 2000)
		Joint laxity	(Baumgartner et al., 2000)
		Cataract	(Baumgartner et al., 2000)
PYCR2	Mutation	Microcephaly	(Escande-Beillard et al., 2020)
		Hypomyelination	(Escande-Beillard et al., 2020)
		Poor overall growth	(Escande-Beillard et al., 2020)
		Delayed psychomotor development	(Escande-Beillard et al., 2020)
	Overexpression	Liver cancer	(Li et al., 2021)
PYCR1	Deletion	ARCL2	(Guernsey et al., 2009)
	Overexpression	Lung cancer	(Li et al., 2021)
		Hepatocellular carcinoma	(Li et al., 2021)
		Colorectal cancer	(Li et al., 2021)
		Gastric cancer	(Li et al., 2021)
		Breast cancer	(Li et al., 2021)
		Prostate cancer	(Li et al., 2021)
		Bladder cancer	(Li et al., 2021)
		Esophagus cancer	(Li et al., 2021)
		Gliomas	(Li et al., 2021)
PRODH	Mutation	Schizophrenia	(de Koning et al., 2015)
		HPI	(Mitsubuchi et al., 2014)
	Reduced expression	Colorectal cancer	(Liu et al., 2009)
P5CDH	Deficiency	HPII	(Mitsubuchi et al., 2014)

cortex of *Prodh* knockout mice (Gogos et al., 1999). In addition, Prodh knockout mice showed a significantly attenuated level of prepulse inhibition, a sensorimotor gating measurement (Gogos et al., 1999). A subsequent study demonstrated that *Prodh* knockout mice exhibited deficits in associative learning and response to psychotomimetic drugs (Paterlini et al., 2005). Furthermore, *Prodh* knockout mice also show that cytosolic accumulation of L-proline disrupts GABAergic transmission through glutamate decarboxylase blockade (Crabtree et al., 2016). In zebrafish, behavioral changes induced by long-term proline exposure are reversed by antipsychotics (Savio et al., 2012), indicating a relationship between proline and psychiatric diseases. *In vitro* studies using differentiated neurons depleted of *Pycr2* showed thinner neuronal fibers and significantly increased axonal beading, an early morphological hallmark of neuronal injury (Escande-Beillard et al., 2020). Collectively, the enzymes in the proline pathway play important roles in neuronal function, brain structure, and behaviors of animals from fish to mammals.

There is accumulating evidence showing that nonneuronal cells, such as astrocytes, play an important role in neurological and psychiatric diseases (Lee et al., 2022; Verkhratsky and Parpura, 2014). An interesting hypothesis for PRODH deficiency leading to hyperactivation of the dopaminergic system is that this is due to dysregulated astroglial control of dopamine homeostasis (de Oliveira et al., 2022). Another opinion is that in the hypothalamus, proline is taken up by astrocytes. Then, proline is converted into lactate, which is then released from astrocytes and taken up by neurons. In neurons, lactate is oxidized for energy production (Arrieta-Cruz and Gutierrez-Juarez, 2016). However, direct evidence is still lacking for a causal role of proline metabolism in nonneuronal cells in the development of neurological and psychiatric diseases.

PROLINE METABOLISM IN OTHER DISEASES

Proline metabolism has been implicated in neurological and psychiatric disorders as well as in other diseases (Table 1). Accumulation of proline can lead to hyperprolinemia due to the dysfunction of catabolic enzymes. There are two types of hyperprolinemia: type I (HPI) and type II (HPII). They are both caused by inborn errors of the proline metabolic pathway (Mitsubuchi et al., 2014). HPI is caused by an abnormality in the PRODH gene, while HPII is caused by a deficiency of the P5CDH gene. The clinical features of HPI are unclear. Nephropathy, uncontrolled seizures, mental retardation and schizophrenia have been reported in HPI, but a benign phenotype without neurological problems has also been reported (Mitsubuchi et al., 2014). PYCRs are also linked to many human diseases. A missense mutation in PYCR1 was found to cause autosomal-recessive cutis laxa type 2 (ARCL2), a multisystem disorder including premature aging, wrinkled and lax skin, joint laxity, and developmental delay (Guernsey et al., 2009). Two homozygous mutations in PYCR2 have been reported to cause microcephaly and hypomyelination, suggesting a crucial role of PYCR2 in the development of the human nervous system (Nakayama et al., 2015).

Dysregulation in proline metabolism is also broadly observed in cancer. PYCRs play an indispensable role in promoting tumorigenesis and cancer progression, and they are

upregulated in many different cancer types, including lung, liver, colorectal, gastric, breast, prostate, bladder, gliomas and esophageal cancer (Li et al., 2021). In breast cancer, PYCR1 but not PYCR2 was also correlated with poor survival (Ding et al., 2017). In contrast, PRODH is a tumor suppressor. PRODH expression levels were much lower in tumor tissues when compared to normal tissues (Liu et al., 2009). When PRODH was inhibited, tumors were readily formed in human colon xenograft animal models. When PRODH was overexpressed, tumors were either suppressed or were so small that the animals did not require euthanasia (Liu et al., 2009).

MOLECULAR EVENTS ASSOCIATED WITH PROLINE METABOLISM

Proline is not only an essential component of proteins but also plays an important role in adaptation to osmotic and dehydration stresses, redox control, and apoptosis. The proline metabolism pathway is conserved in a variety of organisms, e.g., from bacteria to plants and humans (Fichman et al., 2015).

Numerous studies have linked proline metabolism with ROS (Liang et al., 2013). On the one hand, proline acts as an ROS scavenger. For example, in cultured skin fibroblasts, exogenous addition of proline diminished singlet oxygen ($^1O_2$) levels (Wondrak et al., 2005). Moreover, proline can protect human skin cells from photoinduced apoptosis, suggesting that proline can suppress photooxidative stress and skin carcinogenesis (Wondrak et al., 2005). In plants, the production of $^1O_2$ in the thylakoids from the cotyledons of Brassica juncea was dramatically suppressed by proline under strong illumination (Alia et al., 1997). On the other hand, proline metabolism may also lead to increased endogenous ROS (Liu et al., 2012; Szabados and Savoure, 2010; Zarse et al., 2012). In mammalian cells, PRODH activity increases mitochondrial ROS production (Liu et al., 2006). PRODH is a p53-inducible gene (Nagano et al., 2017). Upregulation of PRODH by p53 increases mitochondrial superoxide (O2 • −) production, leading to cytochrome c release and caspase-9 activation (Hu et al., 2007). Elevated ROS levels induced by PRODH have also been associated with the activation of apoptotic pathways via the Ca^{2+}/calcineurin-NFAT cascade (Liu et al., 2006). PRODH is activated by peroxisome proliferator-activated receptor γ (PPARγ), where PRODH-dependent ROS are important mediators of apoptosis in cancer cells treated with PPARγ ligands (Pandhare et al., 2006; Zou et al., 2007). Overall, proline and proline metabolism can act as both an ROS scavenger and producer. Therefore, it is critical to balance proline levels and proline metabolic enzyme activities to achieve proline homeostasis for proper cellular functions.

Proline metabolism may also play a role in the regulation of cellular senescence through ROS and oxidative damage (Campisi, 2013). Inhibition of PRODH expression and/or activity by genetic or pharmacological approaches has been reported to suppress DNA damage-induced senescent phenotypes (Nagano et al., 2017). In contrast, ectopic expression of wild-type PRODH induces senescence, followed by an increase in ROS and an accumulation of DNA damage (Nagano et al., 2017). On the other hand, the proline synthesis enzyme PYCR1 plays an opposite role in cellular senescence. Knockout of *pycr1* in zebrafish resulted in a significant elevation of apoptosis and senescence. Adult *pycr1* knockout fish also exhibited extracellular matrix contents, lowered energy, and diminished superoxide dismutase and telomerase activity when compared to wild-type fish, suggesting that *pycr1* knockout fish may have mitochondrial dysfunctions (Liang et al., 2019). In general, overexpression of PRODH and depletion of PYCR1 lead to increased senescence.

Accelerated senescence has been detected in patients with schizophrenia, AD and PD (Martinez-Cue and Rueda, 2020; Solana et al., 2018). It is plausible that proline metabolism-associated senescence may play a role in schizophrenia, AD and PD. Unlike apoptosis, cellular senescence is typically defined as a state of irreversible cell cycle arrest that releases signaling factors, including proinflammatory cytokines, chemokines, and extracellular matrix proteases, termed the senescence-associated secretory phenotype (SASP) (Fielder et al., 2017). The SASP can be detected by the immune system to restore and maintain organismal health. Both ROS signaling and cellular senescence can lead to cellular immunity. The transcription factor c-Myc triggers the expression of the proline synthesis enzymes PYCR1 and ALDH18A1 in activated T lymphocytes. Additional lines of evidence suggest that proline metabolism contributes to the reprogramming of tumor-infiltrating macrophages (Abuawad et al., 2020; Kuo et al., 2020; Wang et al., 2011). Lon, a mitochondrial chaperone that interacts with PYCR1 in cancer cells, increases intracellular ROS production and activates a p38-NF-κB signaling axis, which in turn stimulates cancer cells to secrete cytokines, including TGF-β, IL-13, IL-6, and VEGF-A, to trigger M2 macrophage polarization (Kuo et al., 2020). Prodh was identified as part of innate immunity by governing ROS homeostasis, and SKN-1 is a critical transcription factor regulating the xenobiotic stress response and pathogen defense activation (Tang and Pang, 2016). In summary, many enzymes in proline metabolism are involved in critical signaling pathways, including ROS, cellular senescence, and cellular immunity, suggesting that proline metabolism may play critical roles in the crosstalk of these cellular processes (Fig. 1).

DISCUSSION

Prolonged disturbances in cell metabolism are likely to lead to neuronal dysfunction, which in turn triggers a series of neurological and psychiatric diseases. PRODH polymorphisms are significantly associated with schizophrenia susceptibility (Guo et al., 2018). There is also evidence that defects in PRODH lead to glutamatergic and GABAergic neuron dysfunctions (Crabtree et al., 2016). However, much remains to be understood. In cancers, it has been proposed that PRODH functions as a tumor suppressor, while PYCRs function as oncogenes. Potential mechanisms include the generation of ROS, triggering cellular senescence and activating the immune system. As ROS signaling, cellular senescence and immunity may also impact the functions of neurons, it is possible that proline metabolism would affect neuronal function through mechanisms such as ROS, cellular senescence and/or cellular immunity. In summary, proline metabolism dysfunction may be a

causal link to neurological or psychiatric diseases by affecting neuronal morphology and function.

In this review, we use proline as an example to demonstrate that amino acid metabolism may play important roles in maintaining normal neuronal functions (Fig. 1). Other metabolic pathways, such as those involved in the metabolism of carbohydrates and lipids, are likely to be equally important. With the rapid development and maturation of analytical tools and cellular preparations, such as single-cell spatial multiomics and human cell-derived organoids, it is hopeful that in-depth understanding of genetic, metabolic and proteomic aspects of cells and the cellular environment will yield exciting insights to shed light on therapeutic strategies for neurological and psychiatric disorders.

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AUTHOR CONTRIBUTIONS

Y.Y. and W.H. conceived and designed the study. Y.Y. performed the literature search and reviewed the previous publications on specific topics. Y.Y. and W.H. wrote the initial draft and reviewed, edited and approved the final version of the submission.

CONFLICT OF INTEREST

The authors have no potential conflicts of interest to disclose.

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REFERENCES

Abuawad, A., Mbadugha, C., Ghaemmaghami, A.M., and Kim, D. (2020). Metabolic characterisation of THP-1 macrophage polarisation using LC–MS-based metabolite profiling. Metabolomics *16*, 33.

Alia, Pardha Saradhi, P., and Mohanty, P. (1997). Involvement of proline in protecting thylakoid membranes against free radical-induced photodamage. J. Photochem. Photobiol. B *38*, 253-257.

Allweis, C., Landau, T., Abeles, M., and Magnes, J. (1966). The oxidation of uniformly labelled albumin-bound palmitic acid to CO2 by the perfused cat brain. J. Neurochem. *13*, 795-804.

Arrieta-Cruz, I. and Gutierrez-Juarez, R. (2016). The role of circulating amino acids in the hypothalamic regulation of liver glucose metabolism. Adv. Nutr. *7*, 790S-797S.

Barber, C.N. and Raben, D.M. (2019). Lipid metabolism crosstalk in the brain: glia and neurons. Front. Cell. Neurosci. *13*, 212.

Bartels, T., De Schepper, S., and Hong, S. (2020). Microglia modulate neurodegeneration in Alzheimer's and Parkinson's diseases. Science *370*, 66-69.

Baumgartner, M.R., Hu, C.A., Almashanu, S., Steel, G., Obie, C., Aral, B., Rabier, D., Kamoun, P., Saudubray, J.M., and Valle, D. (2000). Hyperammonemia with reduced ornithine, citrulline, arginine and proline: a new inborn error caused by a mutation in the gene encoding delta(1)-pyrroline-5-carboxylate synthase. Hum. Mol. Genet. *9*, 2853-2858.

Belanger, M., Allaman, I., and Magistretti, P.J. (2011). Brain energy metabolism: focus on astrocyte-neuron metabolic cooperation. Cell Metab. *14*, 724-738.

Bellon, A. (2007). New genes associated with schizophrenia in neurite formation: a review of cell culture experiments. Mol. Psychiatry *12*, 620-629.

Bender, H.U., Almashanu, S., Steel, G., Hu, C.A., Lin, W.W., Willis, A., Pulver, A., and Valle, D. (2005). Functional consequences of PRODH missense mutations. Am. J. Hum. Genet. *76*, 409-420.

Calabresi, P., Castrioto, A., Di Filippo, M., and Picconi, B. (2013). New experimental and clinical links between the hippocampus and the dopaminergic system in Parkinson's disease. Lancet Neurol. *12*, 811-821.

Campisi, J. (2013). Aging, cellular senescence, and cancer. Annu. Rev. Physiol. *75*, 685-705.

Chan, D.C. (2020). Mitochondrial dynamics and its involvement in disease. Annu. Rev. Pathol. *15*, 235-259.

Clark, J.A. and Amara, S.G. (1993). Amino acid neurotransmitter transporters: structure, function, and molecular diversity. Bioessays *15*, 323-332.

Coyle, J.T. (2006). Glutamate and schizophrenia: beyond the dopamine hypothesis. Cell. Mol. Neurobiol. *26*, 365-384.

Crabtree, G.W., Park, A.J., Gordon, J.A., and Gogos, J.A. (2016). Cytosolic accumulation of L-proline disrupts GABA-ergic transmission through GAD blockade. Cell Rep. *17*, 570-582.

D'Aniello, S., Somorjai, I., Garcia-Fernandez, J., Topo, E., and D'Aniello, A. (2011). D-Aspartic acid is a novel endogenous neurotransmitter. FASEB J. *25*, 1014-1027.

de Koning, M.B., van Duin, E.D., Boot, E., Bloemen, O.J., Bakker, J.A., Abel, K.M., and van Amelsvoort, T.A. (2015). PRODH rs450046 and proline x COMT Val(1)(5)(8) Met interaction effects on intelligence and startle in adults with 22q11 deletion syndrome. Psychopharmacology (Berl.) *232*, 3111-3122.

de Oliveira Figueiredo, E.C., Bondiolotti, B.M., Laugeray, A., and Bezzi, P. (2022). Synaptic plasticity dysfunctions in the pathophysiology of 22q11 deletion syndrome: is there a role for astrocytes? Int. J. Mol. Sci. *23*, 4412.

Dhopeshwarkar, G.A. and Mead, J.F. (1970). Fatty acid uptake by the brain. 3. Incorporation of (1-14C)oleic acid into the adult rat brain. Biochim. Biophys. Acta *210*, 250-256.

Di Paolo, G., Moskowitz, H.S., Gipson, K., Wenk, M.R., Voronov, S., Obayashi, M., Flavell, R., Fitzsimonds, R.M., Ryan, T.A., and De Camilli, P. (2004). Impaired PtdIns(4,5)P2 synthesis in nerve terminals produces defects in synaptic vesicle trafficking. Nature *431*, 415-422.

Dienel, G.A. (2019). Brain glucose metabolism: integration of energetics with function. Physiol. Rev. *99*, 949-1045.

Ding, J., Kuo, M.L., Su, L., Xue, L., Luh, F., Zhang, H., Wang, J., Lin, T.G., Zhang, K., Chu, P., et al. (2017). Human mitochondrial pyrroline-5-carboxylate reductase 1 promotes invasiveness and impacts survival in breast cancers. Carcinogenesis *38*, 519-531.

Ebert, D., Haller, R.G., and Walton, M.E. (2003). Energy contribution of octanoate to intact rat brain metabolism measured by 13C nuclear magnetic resonance spectroscopy. J. Neurosci. *23*, 5928-5935.

Eggers, A.E. (2013). A serotonin hypothesis of schizophrenia. Med. Hypotheses *80*, 791-794.

Escande-Beillard, N., Loh, A., Saleem, S.N., Kanata, K., Hashimoto, Y., Altunoglu, U., Metoska, A., Grandjean, J., Ng, F.M., Pomp, O., et al. (2020). Loss of PYCR2 causes neurodegeneration by increasing cerebral glycine levels via SHMT2. Neuron *107*, 82-94.e6.

Falabella, M., Vernon, H.J., Hanna, M.G., Claypool, S.M., and Pitceathly, R. (2021). Cardiolipin, mitochondria, and neurological disease. Trends Endocrinol. Metab. *32*, 224-237.

Feigenson, K.A., Kusnecov, A.W., and Silverstein, S.M. (2014). Inflammation and the two-hit hypothesis of schizophrenia. Neurosci. Biobehav. Rev. *38*,

72-93.

Fichman, Y., Gerdes, S.Y., Kovacs, H., Szabados, L., Zilberstein, A., and Csonka, L.N. (2015). Evolution of proline biosynthesis: enzymology, bioinformatics, genetics, and transcriptional regulation. Biol. Rev. Camb. Philos. Soc. *90*, 1065-1099.

Fielder, E., von Zglinicki, T., and Jurk, D. (2017). The DNA damage response in neurons: die by apoptosis or survive in a senescence-like state? J. Alzheimers Dis. *60*(s1), S107-S131.

Frajerman, A., Scoriels, L., Kebir, O., and Chaumette, B. (2021). Shared biological pathways between antipsychotics and omega-3 fatty acids: a key feature for schizophrenia preventive treatment? Int. J. Mol. Sci. *22*, 6881.

Franco, R., Rivas-Santisteban, R., Navarro, G., Pinna, A., and Reyes-Resina, I. (2021). Genes implicated in familial Parkinson's disease provide a dual picture of nigral dopaminergic neurodegeneration with mitochondria taking center stage. Int. J. Mol. Sci. *22*, 4643.

GBD 2017 Disease and Injury Incidence and Prevalence Collaborators (2018). Global, regional, and national incidence, prevalence, and years lived with disability for 354 diseases and injuries for 195 countries and territories, 1990-2017: a systematic analysis for the Global Burden of Disease Study 2017. Lancet *392*, 1789-1858.

Gogos, J.A., Santha, M., Takacs, Z., Beck, K.D., Luine, V., Lucas, L.R., Nadler, J.V., and Karayiorgou, M. (1999). The gene encoding proline dehydrogenase modulates sensorimotor gating in mice. Nat. Genet. *21*, 434-439.

Guernsey, D.L., Jiang, H., Evans, S.C., Ferguson, M., Matsuoka, M., Nightingale, M., Rideout, A.L., Provost, S., Bedard, K., Orr, A., et al. (2009). Mutation in pyrroline-5-carboxylate reductase 1 gene in families with cutis laxa type 2. Am. J. Hum. Genet. *85*, 120-129.

Guo, X., Tang, P., Yang, C., and Li, R. (2018). Proline dehydrogenase gene (PRODH) polymorphisms and schizophrenia susceptibility: a meta-analysis. Metab. Brain Dis. *33*, 89-97.

Howes, O.D. and Kapur, S. (2009). The dopamine hypothesis of schizophrenia: version III--the final common pathway. Schizophr. Bull. *35*, 549-562.

Hsieh, H., Boehm, J., Sato, C., Iwatsubo, T., Tomita, T., Sisodia, S., and Malinow, R. (2006). AMPAR removal underlies Abeta-induced synaptic depression and dendritic spine loss. Neuron *52*, 831-843.

Hu, C.A., Donald, S.P., Yu, J., Lin, W.W., Liu, Z., Steel, G., Obie, C., Valle, D., and Phang, J.M. (2007). Overexpression of proline oxidase induces proline-dependent and mitochondria-mediated apoptosis. Mol. Cell. Biochem. *295*, 85-92.

Iwamoto, K., Bundo, M., and Kato, T. (2005). Altered expression of mitochondria-related genes in postmortem brains of patients with bipolar disorder or schizophrenia, as revealed by large-scale DNA microarray analysis. Hum. Mol. Genet. *14*, 241-253.

Kori, M., Aydin, B., Unal, S., Arga, K.Y., and Kazan, D. (2016). Metabolic biomarkers and neurodegeneration: a pathway enrichment analysis of Alzheimer's disease, Parkinson's disease, and amyotrophic lateral sclerosis. OMICS *20*, 645-661.

Kuo, C.L., Chou, H.Y., Chiu, Y.C., Cheng, A.N., Fan, C.C., Chang, Y.N., Chen, C.H., Jiang, S.S., Chen, N.J., and Lee, A.Y. (2020). Mitochondrial oxidative stress by Lon-PYCR1 maintains an immunosuppressive tumor microenvironment that promotes cancer progression and metastasis. Cancer Lett. *474*, 138-150.

Kupeli, A.E., Tatli, C.I., Seker, K.G., Carpar, E., Sobarzo-Sanchez, E., and Capasso, R. (2021). Natural compounds as medical strategies in the prevention and treatment of psychiatric disorders seen in neurological diseases. Front. Pharmacol. *12*, 669638.

Lee, H.G., Wheeler, M.A., and Quintana, F.J. (2022). Function and therapeutic value of astrocytes in neurological diseases. Nat. Rev. Drug Discov. *21*, 339-358.

Lee, K.W., Kim, S.J., Park, J.B., and Lee, K.J. (2011). Relationship between depression anxiety stress scale (DASS) and urinary hydroxyproline and proline concentrations in hospital workers. J. Prev. Med. Public Health *44*, 9-13.

Li, Y., Bie, J., Song, C., Liu, M., and Luo, J. (2021). PYCR, a key enzyme in proline metabolism, functions in tumorigenesis. Amino Acids *53*, 1841-1850.

Liang, S.T., Audira, G., Juniardi, S., Chen, J.R., Lai, Y.H., Du, Z.C., Lin, D.S., and Hsiao, C.D. (2019). Zebrafish carrying pycr1 Gene deficiency display aging and multiple behavioral abnormalities. Cells *8*, 453.

Liang, X., Zhang, L., Natarajan, S.K., and Becker, D.F. (2013). Proline mechanisms of stress survival. Antioxid. Redox Signal. *19*, 998-1011.

Liu, W., Le, A., Hancock, C., Lane, A.N., Dang, C.V., Fan, T.W., and Phang, J.M. (2012). Reprogramming of proline and glutamine metabolism contributes to the proliferative and metabolic responses regulated by oncogenic transcription factor c-MYC. Proc. Natl. Acad. Sci. U. S. A. *109*, 8983-8988.

Liu, Y., Borchert, G.L., Donald, S.P., Diwan, B.A., Anver, M., and Phang, J.M. (2009). Proline oxidase functions as a mitochondrial tumor suppressor in human cancers. Cancer Res. *69*, 6414-6422.

Liu, Y., Borchert, G.L., Surazynski, A., Hu, C.A., and Phang, J.M. (2006). Proline oxidase activates both intrinsic and extrinsic pathways for apoptosis: the role of ROS/superoxides, NFAT and MEK/ERK signaling. Oncogene *25*, 5640-5647.

Martinat, M., Rossitto, M., Di Miceli, M., and Laye, S. (2021). Perinatal dietary polyunsaturated fatty acids in brain development, role in neurodevelopmental disorders. Nutrients *13*, 1185.

Martinez-Cue, C. and Rueda, N. (2020). Cellular senescence in neurodegenerative diseases. Front. Cell. Neurosci. *14*, 16.

Mattson, M.P., Gleichmann, M., and Cheng, A. (2008). Mitochondria in neuroplasticity and neurological disorders. Neuron *60*, 748-766.

Mayneris-Perxachs, J., Castells-Nobau, A., Arnoriaga-Rodriguez, M., Martin, M., de la Vega-Correa, L., Zapata, C., Burokas, A., Blasco, G., Coll, C., Escrichs, A., et al. (2022). Microbiota alterations in proline metabolism impact depression. Cell Metab. *34*, 681-701.e10.

Mitsubuchi, H., Nakamura, K., Matsumoto, S., and Endo, F. (2014). Biochemical and clinical features of hereditary hyperprolinemia. Pediatr. Int. *56*, 492-496.

Nagano, T., Nakashima, A., Onishi, K., Kawai, K., Awai, Y., Kinugasa, M., Iwasaki, T., Kikkawa, U., and Kamada, S. (2017). Proline dehydrogenase promotes senescence through the generation of reactive oxygen species. J. Cell Sci. *130*, 1413-1420.

Nakayama, T., Al-Maawali, A., El-Quessny, M., Rajab, A., Khalil, S., Stoler, J.M., Tan, W.H., Nasir, R., Schmitz-Abe, K., Hill, R.S., et al. (2015). Mutations in PYCR2, encoding pyrroline-5-carboxylate reductase 2, cause microcephaly and hypomyelination. Am. J. Hum. Genet. *96*, 709-719.

Ota, V.K., Bellucco, F.T., Gadelha, A., Santoro, M.L., Noto, C., Christofolini, D.M., Assuncao, I.B., Yamada, K.M., Ribeiro-dos-Santos, A.K., Santos, S., et al. (2014). PRODH polymorphisms, cortical volumes and thickness in schizophrenia. PLoS One *9*, e87686.

Owen, M.J., Sawa, A., and Mortensen, P.B. (2016). Schizophrenia. Lancet *388*, 86-97.

Palop, J.J. and Mucke, L. (2010). Amyloid-beta-induced neuronal dysfunction in Alzheimer's disease: from synapses toward neural networks. Nat. Neurosci. *13*, 812-818.

Pandhare, J., Cooper, S.K., and Phang, J.M. (2006). Proline oxidase, a proapoptotic gene, is induced by troglitazone: evidence for both peroxisome proliferator-activated receptor gamma-dependent and -independent mechanisms. J. Biol. Chem. *281*, 2044-2052.

Paterlini, M., Zakharenko, S.S., Lai, W.S., Qin, J., Zhang, H., Mukai, J., Westphal, K.G., Olivier, B., Sulzer, D., Pavlidis, P., et al. (2005). Transcriptional

and behavioral interaction between 22q11.2 orthologs modulates schizophrenia-related phenotypes in mice. Nat. Neurosci. *8*, 1586-1594.

Phang, J.M., Liu, W., and Hancock, C. (2013). Bridging epigenetics and metabolism: role of non-essential amino acids. Epigenetics *8*, 231-236.

Poewe, W., Seppi, K., Tanner, C.M., Halliday, G.M., Brundin, P., Volkmann, J., Schrag, A.E., and Lang, A.E. (2017). Parkinson disease. Nat. Rev. Dis. Primers *3*, 17013.

Querfurth, H.W. and LaFerla, F.M. (2010). Alzheimer's disease. N. Engl. J. Med. *362*, 329-344.

Rohrbough, J., Rushton, E., Palanker, L., Woodruff, E., Matthies, H.J., Acharya, U., Acharya, J.K., and Broadie, K. (2004). Ceramidase regulates synaptic vesicle exocytosis and trafficking. J. Neurosci. *24*, 7789-7803.

Savio, L.E.B., Vuaden, F.C., Piato, A.L., Bonan, C.D., and Wyse, A.T.S. (2012). Behavioral changes induced by long-term proline exposure are reversed by antipsychotics in zebrafish. Prog. Neuropsychopharmacol. Biol. Psychiatry *36*, 258-263.

Shankar, G.M., Bloodgood, B.L., Townsend, M., Walsh, D.M., Selkoe, D.J., and Sabatini, B.L. (2007). Natural oligomers of the Alzheimer amyloid-beta protein induce reversible synapse loss by modulating an NMDA-type glutamate receptor-dependent signaling pathway. J. Neurosci. *27*, 2866-2875.

Solana, C., Pereira, D., and Tarazona, R. (2018). Early senescence and leukocyte telomere shortening in SCHIZOPHRENIA: a role for cytomegalovirus infection? Brain Sci. *8*, 188.

Sreedharan, J., Blair, I.P., Tripathi, V.B., Hu, X., Vance, C., Rogelj, B., Ackerley, S., Durnall, J.C., Williams, K.L., Buratti, E., et al. (2008). TDP-43 mutations in familial and sporadic amyotrophic lateral sclerosis. Science *319*, 1668-1672.

Srivastava, R., Faust, T., Ramos, A., Ishizuka, K., and Sawa, A. (2018). Dynamic changes of the mitochondria in psychiatric illnesses: new mechanistic insights from human neuronal models. Biol. Psychiatry *83*, 751-760.

Steinlin, M., Boltshauser, E., Steinmann, B., Wichmann, W., and Niemeyer, G. (1989). Hyperprolinaemia type I and white matter disease: coincidence or causal relationship? Eur. J. Pediatr. *149*, 40-42.

Stokin, G.B. and Goldstein, L.S. (2006). Axonal transport and Alzheimer's disease. Annu. Rev. Biochem. *75*, 607-627.

Surmeier, D.J. (2007). Calcium, ageing, and neuronal vulnerability in Parkinson's disease. Lancet Neurol. *6*, 933-938.

Sweeney, M.D., Zhao, Z., Montagne, A., Nelson, A.R., and Zlokovic, B.V. (2019). Blood-brain barrier: from physiology to disease and back. Physiol. Rev. *99*, 21-78.

Szabados, L. and Savoure, A. (2010). Proline: a multifunctional amino acid. Trends Plant Sci. *15*, 89-97.

Tang, H. and Pang, S. (2016). Proline catabolism modulates innate immunity in Caenorhabditis elegans. Cell Rep. *17*, 2837-2844.

Travagli, R.A., Browning, K.N., and Camilleri, M. (2020). Parkinson disease and the gut: new insights into pathogenesis and clinical relevance. Nat. Rev. Gastroenterol. Hepatol. *17*, 673-685.

Uno, Y. and Coyle, J.T. (2019). Glutamate hypothesis in schizophrenia. Psychiatry Clin. Neurosci. *73*, 204-215.

Verkhratsky, A. and Parpura, V. (2014). Neurological and psychiatric disorders as a neuroglial failure. Period. Biol. *116*, 115-124.

Wang, R., Dillon, C.P., Shi, L.Z., Milasta, S., Carter, R., Finkelstein, D., McCormick, L.L., Fitzgerald, P., Chi, H., Munger, J., et al. (2011). The transcription factor Myc controls metabolic reprogramming upon T lymphocyte activation. Immunity *35*, 871-882.

Wondrak, G.T., Jacobson, M.K., and Jacobson, E.L. (2005). Identification of quenchers of photoexcited States as novel agents for skin photoprotection. J. Pharmacol. Exp. Ther. *312*, 482-491.

Xie, K., Qin, Q., Long, Z., Yang, Y., Peng, C., Xi, C., Li, L., Wu, Z., Daria, V., Zhao, Y., et al. (2021). High-throughput metabolomics for discovering potential biomarkers and identifying metabolic mechanisms in aging and Alzheimer's disease. Front. Cell Dev. Biol. *9*, 602887.

Zarse, K., Schmeisser, S., Groth, M., Priebe, S., Beuster, G., Kuhlow, D., Guthke, R., Platzer, M., Kahn, C.R., and Ristow, M. (2012). Impaired insulin/IGF1 signaling extends life span by promoting mitochondrial L-proline catabolism to induce a transient ROS signal. Cell Metab. *15*, 451-465.

Zielke, H.R., Zielke, C.L., and Baab, P.J. (2009). Direct measurement of oxidative metabolism in the living brain by microdialysis: a review. J. Neurochem. *109* Suppl 1, 24-29.

Zou, W., Liu, X., Yue, P., Khuri, F.R., and Sun, S.Y. (2007). PPARgamma ligands enhance TRAIL-induced apoptosis through DR5 upregulation and c-FLIP downregulation in human lung cancer cells. Cancer Biol. Ther. *6*, 99-106.