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The Key Role of NADPH in Biosynthesis and Antioxidant Reactions

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Abstract NADPH, the full name of which is nicotinamide adenine dinucleotide phosphate, is a very important coenzyme in cells. It plays a key role in the body's production of various substances and resistance to oxidative stress. We can think of it as an "electron provider" that is indispensable in many biological reactions. When synthesizing fatty acids, cholesterol, nucleotides, and some secondary metabolites, cells need a lot of NADPH to provide reducing power. In other words, without it, these things cannot be made. NADPH also helps cells fight "oxidative stress". It can regenerate reduced glutathione and support the normal operation of the thioredoxin system. These systems are particularly important for scavenging free radicals and maintaining cell health. The main way cells make NADPH is through a metabolic pathway called the "pentose phosphate pathway". In addition, some enzymes, such as NADP-dependent malic enzyme and isocitrate dehydrogenase, can also replenish NADPH. These enzymes are distributed in different areas of the cell to ensure that there is enough NADPH everywhere. NADPH is also important in the immune system. For example, when phagocytes (like macrophages) react in an outburst, it helps cells quickly release reactive substances to kill bacteria. In addition, NADPH is also related to aging, metabolic problems, and the development of certain diseases. Scientists are studying how to better monitor and regulate NADPH in cells. These new technologies may be used in medicine, agriculture, and biotechnology. Future research needs to further clarify how NADPH is regulated and how it interacts with various cell signaling pathways. NADPH is critical for maintaining cell health and may also become a new target for treating certain diseases (especially those related to oxidative stress) or improving metabolic efficiency.
Keywords NADPH; Biosynthesis; Antioxidant reactions; Redox homeostasis; Metabolic regulation

1 Introduction

NADPH, the full name of which is nicotinamide adenine dinucleotide phosphate, is a very important coenzyme in cell metabolism. It plays a key role in redox reactions. Structurally, NADPH and NADH are very similar. The main difference between them is that there is an extra phosphate on the ribose ring of NADPH. This small change allows NADPH to specifically participate in anabolism. It can provide the required reducing power in the synthesis of substances and antioxidant defense (Xiao et al., 2018; Amjad et al., 2021). NADPH does not have only one source. It can be synthesized through several different metabolic methods. For example, the oxidative pentose phosphate pathway and a step in the tricarboxylic acid cycle, isocitrate dehydrogenase, can also help produce NADPH (Spaans et al., 2015). NADPH is indispensable in many life activities, especially in the manufacture of substances and anti-oxidation. It provides reducing power in the synthesis of fatty acids, cholesterol and nucleotides. These substances are very important for cell growth and division (Xiao et al., 2018; Amjad et al., 2021). NADPH also helps cells resist oxidative stress. It can restore the antioxidant glutathione to an active state. This process helps cells fight damage caused by reactive oxygen species (ROS) (Hashida et al., 2010; Spaans et al., 2015). Because of these functions, NADPH is particularly important in cellular metabolism. It is also considered a new target for treating metabolic diseases and some degenerative diseases (Braidy et al., 2019; Tannous et al., 2020).

This study mainly wants to take a deeper look at the role of NADPH in material production and anti-oxidation. We will start with its molecular structure, then talk about how it is made, and analyze what it does specifically in cellular activities. Our goal is to fully understand the role of NADPH in supporting cellular metabolism and think

about its possible future application in treating diseases. By organizing and analyzing the current research results, we hope to further illustrate the importance of NADPH to cellular health and explore its potential as a therapeutic tool.

2 NADPH and Cellular Metabolism

2.1 Detailed examination of NADPH's generation pathways

NADPH is produced by several metabolic pathways in cells, the most important of which is the pentose phosphate pathway (PPP), and some enzymes, such as malic enzyme, can also participate. The oxidation phase of PPP is critical and is mainly completed by two enzymes: glucose-6-phosphate dehydrogenase (G6PDH) and 6-phosphogluconate dehydrogenase (6PGDH). These two enzymes help cells produce NADPH (Corpas et al., 2020; Fuentes-Lemus et al., 2023). In addition to PPP, there are other enzymes that can also produce NADPH, such as NADP-dependent malic enzyme (NADP-ME) and NADP-dependent isocitrate dehydrogenase (NADP-ICDH). These enzymes are distributed in different areas of the cell, such as the cytoplasm, mitochondria and peroxisomes (Corpas and Barroso, 2014; Corpas et al., 2020). It is precisely because of their existence that NADPH can be stably supplied in the cell, which is important for various anabolic and detoxification reactions.

2.2 NADPH's role in maintaining cellular redox balance

NADPH is active in many antioxidant systems of the cell, where it is the main reductant and helps the cell maintain a normal redox state. NADPH is important for the action of glutathione reductase, an enzyme that regenerates reduced glutathione (GSH), which is critical for the cell to resist oxidative damage (Lee et al., 2002). NADPH also participates in the ascorbate-glutathione cycle and supports the activity of NADPH oxidase. This oxidase generates reactive oxygen species (ROS), which can be used for cell signaling (Corpas et al., 2020; Fuentes-Lemus et al., 2023). NADPH also contributes to the normal function of thioredoxin reductase, an enzyme that regulates the status of thioredoxin. Thioredoxin itself is also an important antioxidant tool (Corpas et al., 2020). The production and use of NADPH must be balanced to prevent oxidative stress and maintain a stable environment in the cell (Ying, 2008).

2.3 Comparative analysis with other cellular cofactors

In cellular metabolism, NADPH, NADH and FADH2 are all very important cofactors, but their respective tasks are different. NADH is mainly involved in catabolism, such as glycolysis and the tricarboxylic acid cycle (TCA), and it can help cells produce ATP through oxidative phosphorylation (Blacker and Duchen, 2016; Xiao et al., 2018). NADPH is more used for anabolism. It can support the synthesis of fatty acids and nucleotides, and also help cells maintain a healthy redox environment (Ying, 2008; Corpas and Barroso, 2014).

FADH2 is similar to NADH and also participates in the electron transport chain, which is important for the synthesis of ATP (Xiao et al., 2018). Although both NADH and FADH2 are more inclined to energy generation, the role of NADPH is more focused on making substances needed by cells and protecting cells from oxidation, which also explains its unique position in metabolism (Blacker and Duchen, 2016; Shimizu and Matsuoka, 2019).

3 Biosynthesis Reactions Facilitated by NADPH

3.1 Lipid biosynthesis: role of NADPH in fatty acid and cholesterol biosynthesis

NADPH is critical in the process of lipid production, especially in the synthesis of fatty acids and cholesterol. $NADP^+$-dependent isocitrate dehydrogenase (IDPc) in the cytoplasm is an important source of NADPH. It plays an important role in these synthesis reactions. When adipocytes differentiate, the activity of IDPc increases and the expression level also increases. This is closely related to the increase in fat formation. Transgenic experiments conducted on mice found that excessive expression of IDPc can lead to fatty liver, increased blood lipids, and obesity. This shows that it plays an important role in lipid metabolism (Koh et al., 2004). NADPH is also very important for the activity of two key enzymes: fatty acid synthase and HMG-CoA reductase. The former is a key enzyme in fatty acid synthesis, and the latter is involved in cholesterol synthesis (Pollak et al., 2007; Spaans et al., 2015).

3.2 Amino acid and nucleotide synthesis: NADPH in the synthesis of non-essential amino acids and nucleotides

NADPH is also indispensable in the synthesis of non-essential amino acids and nucleotides. For example, the synthesis of proline requires NADPH in mitochondria. NAD kinase 2 (NADK2) can generate mitochondrial $NADP^+$. If there is a problem in this process, it will lead to insufficient NADPH in mitochondria, which will affect the production of proline. This shows that mitochondrial NADPH is indispensable in the synthesis of pyrroline-5-carboxylate, an intermediate in the synthesis of proline (Tran et al., 2021). In addition to amino acids, NADPH is also involved in the production of nucleotides. In the pentose phosphate pathway (PPP), NADPH helps synthesize ribose-5-phosphate, a sugar that is a raw material for nucleotides. At the same time, NADPH also provides reducing power (Corpas and Barroso, 2014; Xiao et al., 2018) (Figure 1).

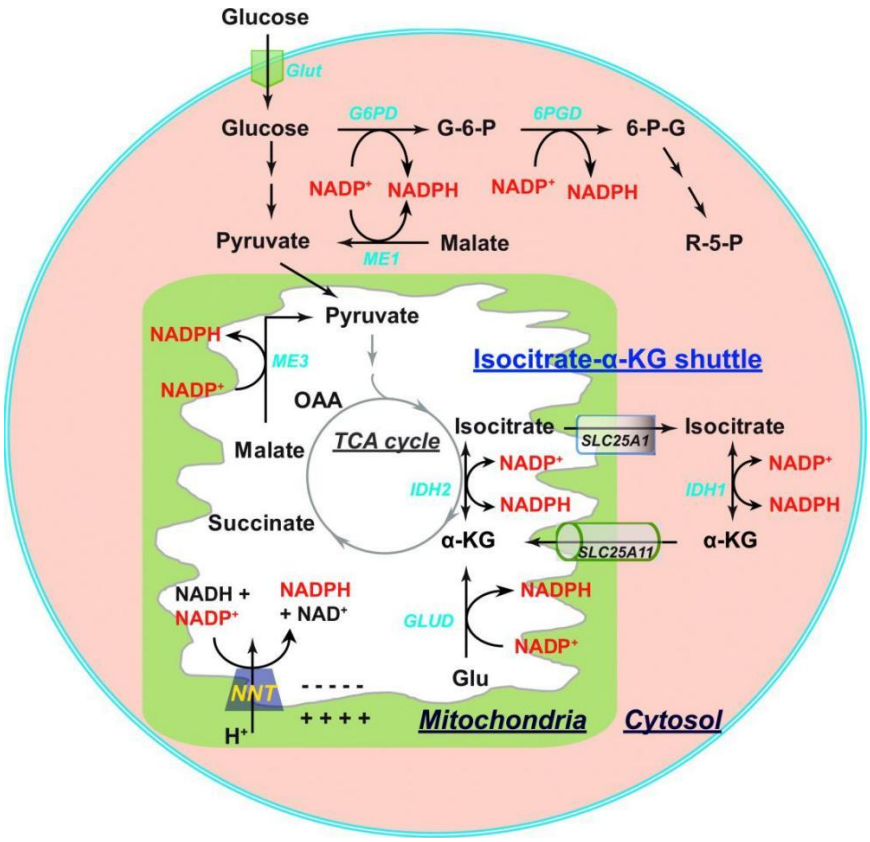

Figure 1 Metabolic sources of NADP(H) and the cytosolic/mitochondrial NADPH shuttle (Adopted from Xiao et al., 2018)

Image caption: In the cytosol, NADPH is primarily produced by G6PD and 6PGD in the pentose phosphate pathway. ME1 also contributes to cytosolic NADPH production. Mitochondrial NADPH is generated by $NADP^+$-dependent IDH2, GLUD, NNT, and ME3. The cytosolic and mitochondrial NADPH is exchanged through the isocitrate-α-KG shuttle, where cytosolic IDH1 and mitochondrial IDH2 catalyze the interconversion of isocitrate and α-KG in conjunction with the interconversion of $NADP^+$ and NADPH. The citrate carrier protein (encoded by *SLC25A1* gene) and the α-KG/malate antiporter (encoded by *SLC25A11* gene) mediate the transport of isocitrate and α-KG between cytosol and mitochondria, respectively. 6PG, 6-phosphogluconate; 6PGD, 6-phosphogluconate dehydrogenase; G6P, glucose-6-phosphate; G6PD, glucose-6-phosphate dehydrogenase; NNT, nicotinamide nucleotide transhydrogenase; R5P, ribose-5-phosphate; SCL25A1, solute carrier family 25 member 1 (Adopted from Xiao et al., 2018)

3.3 Secondary metabolite synthesis: importance of NADPH in the synthesis of plant secondary metabolites and pharmaceuticals

NADPH is critical for plants to produce certain special compounds, which can often be used as drugs. NADPH provides reducing power to the enzyme reactions that synthesize these molecules. Enzymes such as cytochrome P450 require NADPH to work properly. This enzyme can hydroxylate some substrates, such as flavonoids, alkaloids, terpenes, etc., which are common plant secondary metabolites (Hajeyah et al., 2020). There are also some NADPH-dependent oxidoreductases, such as ferredoxin-NDP reductase and NADP-dependent malic

enzyme, which also help maintain NADPH levels in cells. The activity of these enzymes can further promote the synthesis of various important metabolites (Corpas et al., 2020). NADPH is not only useful in basic metabolism, but also very important in these more complex secondary metabolic pathways. It helps synthesize many biologically active substances, such as plant medicinal ingredients (Takayanagi et al., 1980; Blacker et al., 2014).

4 NADPH in Antioxidant Defense Mechanisms

4.1 Overview of oxidative stress and the necessity of antioxidants in cellular protection

Oxidative stress occurs because there are too many reactive oxygen species (ROS) and the cells are not able to remove these harmful molecules. ROS include free radicals and peroxides, which are byproducts of normal cell metabolism, especially aerobic respiration. If too much ROS is produced, it will damage the DNA, proteins and fats in the cell. This will impair cell function and even cause cell death. In order to cope with this stress, cells have developed many antioxidant systems. These systems include enzymes and non-enzymatic antioxidants, which together help cells maintain redox balance and prevent damage (Benhar, 2018; Hasan et al., 2022; Chai and Mieyal, 2023).

4.2 Glutathione redox cycle: NADPH's role in regenerating reduced glutathione

The glutathione (GSH) redox cycle is an important antioxidant mechanism in cells. GSH is a common non-enzymatic antioxidant that can directly remove ROS. In this process, GSH will become oxidized GSSG. At this time, the cell needs to use NADPH and glutathione reductase to convert GSSG back to GSH. This process is particularly critical for maintaining the redox balance of cells and can also help cells fight oxidative stress. The GSH/Grx system can also regulate the S-glutathionylation reaction of proteins. This process is reversible and also requires the participation of NADPH. It is very helpful in maintaining the signal transduction and red oxygen status of cells (Bradshaw, 2019; Ferguson and Bridge, 2019; Chai and Mieyal, 2023).

4.3 Thioredoxin system: function of NADPH in the thioredoxin system and its significance in DNA repair

The thioredoxin (Trx) system is another important antioxidant system. It consists of Trx, Trx reductase (TrxR) and NADPH. The role of Trx is to help proteins restore their structure, and it can reduce disulfide bonds in proteins. The function of TrxR is to use NADPH to reduce oxidized Trx. This process not only helps proteins maintain function, but is also closely related to DNA repair. Because many proteins involved in DNA synthesis and repair also need to maintain the correct redox state. This Trx/TrxR system also plays a role in cell proliferation and survival, so it is very important for the entire cell homeostasis (Benhar, 2018; Ferguson and Bridge, 2019; Hasan et al., 2022).

4.4 Antioxidant enzymes: interaction of NADPH with enzymes like catalase and superoxide dismutase in reducing oxidative damage

NADPH is also involved in supporting the function of several antioxidant enzymes, such as catalase and superoxide dismutase (SOD). Catalase can break down harmful hydrogen peroxide into water and oxygen, while SOD can turn superoxide free radicals into less dangerous substances. These enzymes will be oxidized after working, and NADPH can provide reducing power to restore them to a state where they can continue to work, so that they can continue to work and protect cells from oxidative damage (Figure 2). NADPH is also involved in the ascorbic acid-glutathione cycle and the NADPH-dependent thioredoxin system. This once again illustrates the central position of NADPH in the cellular antioxidant system (Ryoo and Kwak, 2018; Corpas et al., 2020; Liu et al., 2020).

5 Regulation of NADPH Homeostasis

5.1 Mechanisms that cells employ to regulate NADPH levels

Cells control NADPH levels in a variety of ways to maintain a balance between its production and use. NADPH is primarily generated through the pentose phosphate pathway (PPP). This pathway is regulated by glucose-6-phosphate dehydrogenase (G6PD) and AMP kinase (Tao et al., 2017). Another way is through NAD^+ kinases in the cytoplasm and mitochondria, which convert NAD^+ to $NADP^+$ and then reduce it to NADPH (Bradshaw, 2019). Cells store NADPH in different areas, such as in the cytoplasm and mitochondria, which helps

maintain redox balance and support various synthetic reactions (Xiao et al., 2018). NADPH oxidase (NOX) is also involved in regulation. It produces reactive oxygen species (ROS) as part of cell signaling and also affects NADPH levels (Ewald, 2018; Ryoo and Kwak, 2018).

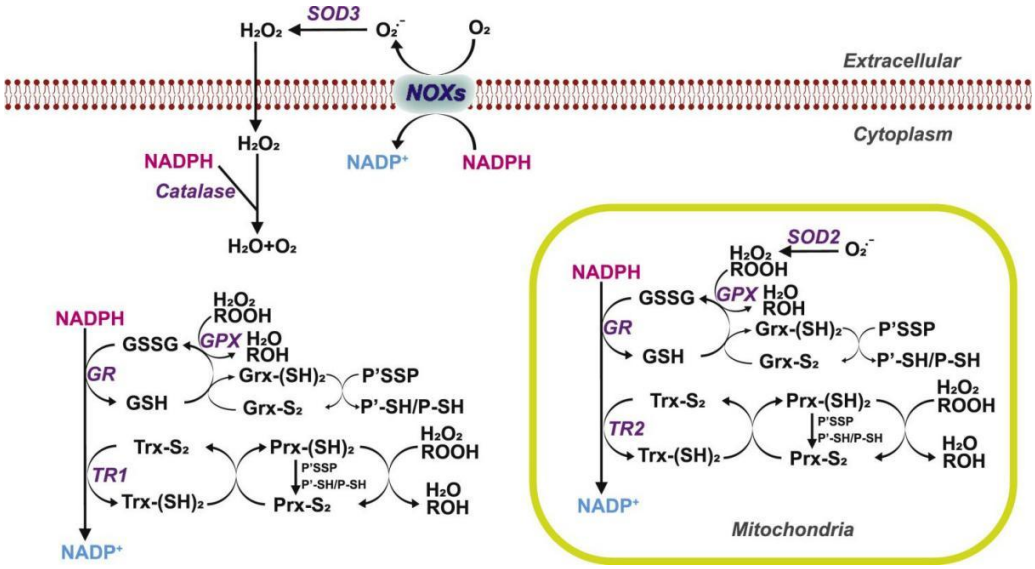

Figure 2 NADPH in regulating cellular redox homeostasis (Adopted from Liu et al., 2020)

Image caption: NADPH is used as a reducing power in GR- or TR1/2-mediated antioxidant reactions. GR reduces GSSG to GSH, which is then used as a co-factor by GPXs, and GSH is re-oxidized to GSSG. In addition, the oxidation of GSH to GSSG is coupled with the reduction of glutaredoxin disulfide Grx-S_2 to glutaredoxin thiol Grx-(SH_2), which can conduct thiol–disulfide exchange reactions with protein disulfides (P'SSP), generating reduced protein thiols (P′-SH/P-SH). Cytosolic TR1 or mitochondrial TR2 catalyzes the reduction of thioredoxin disulfide Trx-S2 to thioredoxin thiols Trx-$(SH)_2$, which in turn promote the reduction of peroxiredoxin disulfide Prx-S2 to peroxiredoxin thiols Prx-$(SH)_2$. Prx-$(SH)_2$ may directly detoxify ROS or reduce protein disulfide by thiol-disulfide exchange. Transmembrane enzymes NOXs primarily catalyze the formation of superoxide anion O_2^-, which is rapidly converted to H_2O_2 by SOD3. In addition, NADPH binds to catalase converting H_2O_2 to H_2O. Proteins are indicated in purple. GR: glutathione reductase; TR1/2: thioredoxin reductase 1/2; SOD2/3: superoxide dismutase 2/3; NOXs: NADPH oxidases; ROOH: alkyl hydroperoxide; ROH: alkyl alcohol; P'SSP: protein disulfide; P′-SH/P-SH: protein thiol (Adopted from Liu et al., 2020)

5.2 Impact of nutrient availability and environmental stress on NADPH production

NADPH production is affected by nutritional conditions and environmental stress. When cells are under oxidative stress, they increase NADPH production to enhance antioxidant capacity. This process is mainly accomplished by activating G6PD in PPP, and this pathway is regulated by both oxidative stress and nutritional signals. In particular, the amount of glucose is very important for NADPH homeostasis. Glucose metabolism can provide NADPH and is its main source. Therefore, when glucose levels fluctuate, NADPH production will also be affected (Tao et al., 2017). Environmental stress, such as hypoxia, can also change the activity of enzymes that produce NADPH or affect redox reactions in cells, thereby affecting NADPH levels (Xiao et al., 2018). These changes allow cells to respond flexibly to external stress and help them survive in adversity (Blacker and Duchen, 2016).

5.3 Role of NADPH in aging and disease

NADPH is important in regulating redox balance and metabolism in cells, and therefore plays a key role in aging and various diseases. NADPH levels tend to decrease with age. This can induce more oxidative stress and cause cell damage. The cause of this decline may be related to the reduced activity of enzymes related to NADPH synthesis and the deterioration of mitochondrial function. In some animal experiments, if cells are allowed to produce more NADPH, such as overexpressing synthases, life span can be extended. This suggests that there may be a positive correlation between NADPH levels and life span (Bradshaw, 2019). In terms of disease, NADPH homeostasis is particularly closely related to cancer. Cancer cells adjust their metabolism and need more NADPH to support antioxidant and anabolic reactions. Therefore, targeting NADPH metabolism in cancer cells may be a

therapeutic idea, and their sensitivity to oxidative stress can be used to fight tumors (Ju et al., 2020). NADPH oxidases are also involved in the occurrence of many diseases, such as cardiovascular disease and neurodegenerative diseases. It participates in the processes of these diseases by producing ROS and regulating signaling pathways (Chan et al., 2009 ; Maraldi et al., 2021).

6 NADPH Deficiency and Its Implications

6.1 Diseases associated with NADPH deficiency

NADPH deficiency is closely related to an immune disease called chronic granulomatous disease (CGD). CGD is a congenital immunodeficiency disease that is mainly caused by malfunction of NADPH oxidase in phagocytes. When this enzyme does not work properly, it cannot produce enough reactive oxygen species (ROS), which are used to kill bacteria and fungi. This makes people more susceptible to repeated infections and the symptoms are more severe (Stasia and Li, 2008; Arnold and Heimall, 2017; Yu et al., 2020). The cause of CGD is usually a genetic mutation in certain subunits of NADPH oxidase, such as CYBB ($gp91^{phox}$), NCF1 ($p47^{phox}$) and CYBA ($p22^{phox}$) (Stasia and Li, 2008; Yu et al., 2020). These patients not only have infection problems, but are also prone to inflammation, such as granulomas or intestinal inflammatory diseases (Arnold and Heimall, 2017; Yu et al., 2020; León-Lara et al., 2021). In addition, some CGD patients and women with X-linked mutations also develop autoimmune diseases (Yu et al., 2020).

6.2 Impact of NADPH levels on immune function and disease progression

NADPH is important for the functioning of the immune system, especially in phagocytes. It helps these cells to perform an "oxidative burst," a step that is necessary to kill pathogens. If NADPH oxidase activity is insufficient, cells will have difficulty clearing bacteria or viruses, making them more susceptible to infection (Hohn and Lehrer, 1975; Giardino et al., 2017; Violi et al., 2017). Not only that, this deficiency can also make inflammation excessive because cells cannot properly regulate and shut down related signaling pathways. Studies have found that lack of NADPH activates the NF-κB pathway and increases pro-inflammatory factors, resulting in chronic inflammation and even tissue damage (Segal et al., 2010; Segal et al., 2012; León-Lara et al., 2021). In addition, some studies have shown that even a small amount of residual activity of NADPH oxidase can affect the severity of the disease and the life expectancy of patients, indicating that maintaining a certain level of NADPH is really important (Stasia and Li, 2008; Yu et al., 2020).

6.3 Therapeutic interventions to enhance NADPH availability

Currently, there are some ways to try to solve the problem of NADPH deficiency. Hematopoietic stem cell transplantation (HSCT) is currently the only cure for CGD, especially for children, with a survival rate of more than 90% (Arnold and Heimall, 2017; Yu et al., 2020). Gene therapy is also a developing direction. This method is to re-implant the repaired hematopoietic stem cells into the body in the hope of restoring the function of NADPH oxidase. Although the effect is not stable enough at present, many clinical trials are being improved in order to achieve a more lasting therapeutic effect (Barese et al., 2004; Yu et al., 2020). In addition to these, there are some drugs under study. These drugs mainly target inflammation or oxidative stress caused by CGD, such as pioglitazone, tamoxifen and rapamycin, which can help control symptoms (Yu et al., 2020; León-Lara et al., 2021). There is also a new class of compounds, such as CDDO-Im (belonging to triterpenes), which can activate anti-inflammatory pathways and are independent of NADPH oxidase. This type of drug shows the potential to reduce inflammation in CGD models (Segal et al., 2010).

7 NADPH in Immune Response and Cellular Defense

7.1 Examination of NADPH's role in immune cell function, particularly in phagocytes

NADPH is very important in immune cells, especially in phagocytes, such as neutrophils and macrophages. It helps these cells produce reactive oxygen species (ROS) to kill invading pathogens. There is an enzyme called NADPH oxidase on the membrane of these cells, which can rapidly release ROS in a respiratory burst. This burst is critical for phagocytes to eliminate the bacteria or fungi they ingest (Thomas, 2017; Moghadam et al., 2021; Vermot et al., 2021). NADPH oxidase is not a single molecule, but a complex composed of several parts. It

includes gp91^{phox} and p22^{phox} on the membrane, as well as p47^{phox}, p67^{phox}, p40^{phox} and Rac1/2 in the cytoplasm. These components assemble when activated and then generate reactive oxygen species (El-Benna et al., 2005; El-Benna et al., 2009; Nunes et al., 2013). The entire process is strictly controlled to avoid excessive reactive oxygen species that damage tissues while ensuring a bactericidal effect (Mcphail et al., 1985; Edwards, 1996) (see Figure 3).

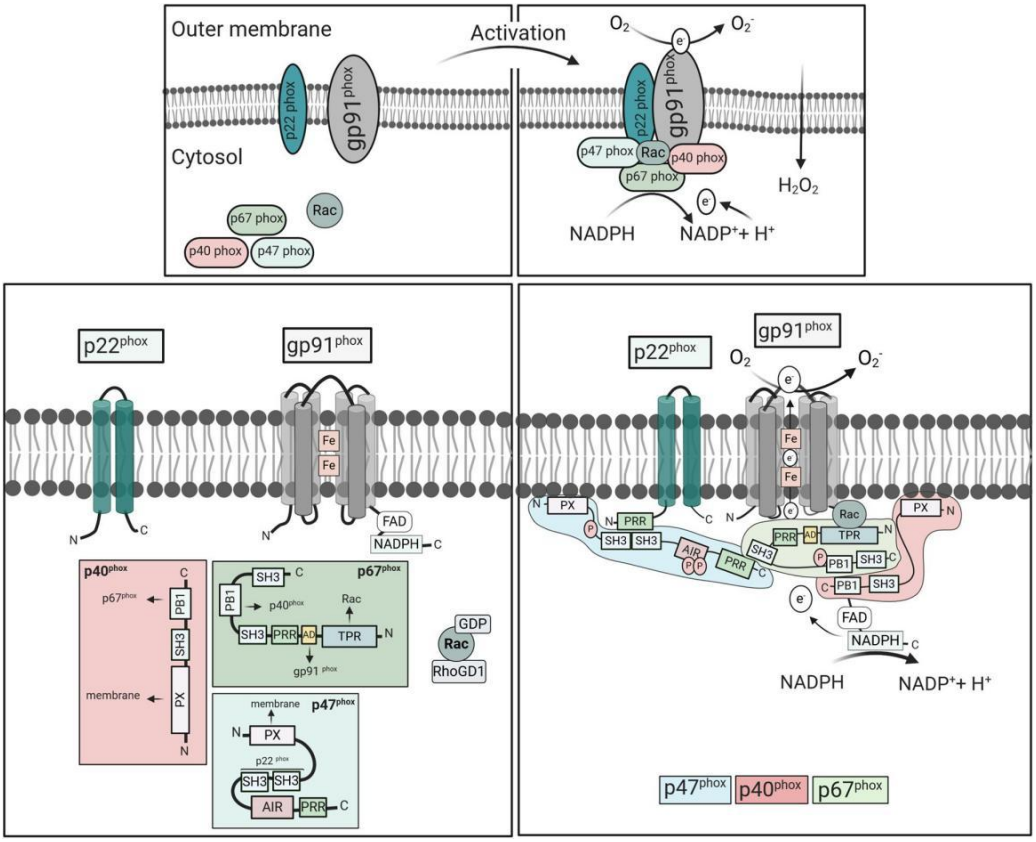

Figure 3 Activation and assembly of mammalian NOX2. NOX2 consists of the cytosolic components p67^{phox}, p47^{phox}, p40^{phox}, Rac2, and the integral membrane subunits gp91^{phox} and p22^{phox}. Upon cell stimulation, the cytosolic subunits translocate to the membranes to form an active complex with gp91^{phox} and p22^{phox}. Meanwhile, Rac exchanges GDP to GTP, and dissociates from Rho-GDI. In the resting state, the p47^{phox}-SH3 tandem domain interacts with AIR keeping p47phox in an inactive conformation (Belambri et al., 2018). Cell stimulation induces phosphorylation of AIR, releasing the interactive domains, i.e., SH3, PX, and PRR, which mediate oxidase assembly. The PRR of p47^{phox} binds to the SH3 region of p67^{phox}, while p67^{phox} links with p40^{phox} through their PB1 domains. The p47^{phox}-SH3 regions then bind to the p22^{phox}-PRR domains promoting p67^{phox} interaction with gp91^{phox} and moving p40^{phox}-PX domains in close proximity to the membrane. Activated NOX2 uses cytosolic NADPH to induce oxygen reduction and superoxide anion (O_2^-) generation. Abbreviations: SH3, Src homology 3 (SH3);, PX, phox homology (PX); AIR, auto-inhibitory region (AIR);, PRR, proline-rich region (PRR);, TPR, tetratricopeptide-rich regions; PB1, phox and Bem1 domain; and AD, activation domain (Adopted from Moghadam et al., 2021)

7.2 Contribution of NADPH to the respiratory burst and microbial defense

The respiratory burst is a fast reaction that rapidly releases a large number of ROS, such as superoxide anions and hydrogen peroxide. These reactive oxygen species are generated by NADPH oxidase. Once NADPH oxidase is activated, it transfers electrons from NADPH to oxygen molecules to produce superoxide anions. These reactive oxygen species are then converted into other ROS (Kotsias et al., 2013; Nunes et al., 2013; Thomas, 2017). These ROS are lethal to the engulfed microorganisms, which can be completely killed in the phagosome. Chronic granulomatous disease illustrates the importance of this process. This disease is caused by a malfunction of part of the NADPH oxidase, which makes it difficult for the patient's immune system to kill bacteria, resulting in repeated infections (El-Benna et al., 2005; Glennon-Alty et al., 2018).

7.3 Implications for immune health and potential targets for enhancing NADPH in immunocompromised individuals

If NADPH oxidase does not work properly, it will make the immune system very vulnerable. For example, in CGD patients, it can be seen that the failure of this enzyme leads to repeated infections and low immune function (El-Benna et al., 2005; Glennon-Alty et al., 2018). To improve this situation, scientists are trying some methods to increase the activity of NADPH oxidase or find other ways to make up for its deficiency. You can try to regulate the activation process of this enzyme. Methods such as phosphorylating $p47^{phox}$ or promoting its binding to PI(3)P may enhance the activity of the enzyme (El-Benna et al., 2009; Nunes et al., 2013). A deeper understanding of the role of reactive oxygen species in immune signaling and tissue regulation may also help us develop new treatments. These methods can effectively kill bacteria while reducing tissue damage (Edwards, 1996; Moghadam et al., 2021; Vermot et al., 2021).

8 Emerging Research and Future Directions

8.1 Recent advances in NADPH-related research

Recent studies have made us more aware of the importance of NADPH in cellular metabolism and redox balance. Now, scientists have developed some new tools, such as genetically encoded biosensors. It can track NADPH levels in different regions of the cell in real time. This helps us understand how cells maintain the distribution of NADPH in different locations to avoid oxidative stress (Xiao et al., 2018). Other studies have found new enzymes that synthesize NADPH, which is very helpful for understanding its production and role in metabolic pathways (Pollak et al., 2007). At the same time, studies have once again emphasized the role of NADPH in clearing oxidative stress, such as helping to regenerate reduced glutathione or participating in other detoxification processes (Lee et al., 2002). A technology called fluorescence lifetime imaging microscopy (FLIM) can now distinguish NADH and NADPH in living cells. This allows us to study their respective metabolic functions from a new perspective (Blacker et al., 2014).

8.2 Potential applications of NADPH modulation in medicine, agriculture, and biotechnology

Manipulating NADPH levels has great potential in medicine, agriculture, and biotechnology. In medicine, adjusting the metabolic pathways of NADPH may help treat diseases related to oxidative stress or metabolic disorders, such as diabetes, neurodegenerative diseases, and cancer (Ying, 2008; Blacker and Duchen, 2016). For example, increasing NADPH production or enhancing its utilization can enhance the ability of cells to resist oxidative damage and support tissue repair (Chan et al., 2009; Henríquez-Olguín et al., 2019). In agriculture, regulating the enzymes that produce NADPH in plants may also enhance the resistance of crops to environmental stresses such as heat and drought. This will not only help increase yields, but also make agriculture more sustainable (Corpas and Barroso, 2014; Corpas et al., 2020). In the field of biotechnology, optimizing the efficiency of NADPH-related reactions can help improve the biosynthetic capacity of industrial strains, such as producing biofuels and other valuable metabolites (Pollak et al., 2007).

8.3 Unresolved questions and future research needs

Although we have made some progress, there are still many questions that need to be further studied. A key question is: How do cells adjust the distribution of NADPH under different physiological or pathological conditions? How do these changes help cells maintain redox balance? This is still unclear (Ying, 2008; Xiao et al., 2018). The connection between NADPH and other signaling pathways, such as hypoxia-inducible factor (HIF) or calcium signaling, also needs further investigation. There may be complex interactions between them, which will affect the cell's stress response and metabolism (Lee et al., 2002; Xiao et al., 2018). We also need to develop more advanced tools to measure or control NADPH levels in vivo. This will help us understand its dynamic changes at different time points and locations (Blacker et al., 2014). If we really use methods to regulate NADPH in clinical or agricultural applications in the future, we must also evaluate its side effects and long-term effects in advance to ensure that these methods are safe and effective (Chan et al., 2009; Henríquez-Olguín et al., 2019).

9 Conclusion

NADPH plays an important role in cellular metabolism. It is a provider of electrons for many anabolic and antioxidant reactions. It provides reducing power for the synthesis of fatty acids, cholesterol, and nucleotides, all of which are important for cells. In addition, NADPH helps regenerate reduced glutathione. This substance is critical for protecting cells from oxidative damage. Although NADPH is primarily produced by the oxidative pentose phosphate pathway (PPP), other pathways are involved, such as the malic enzyme pathway and folate-related one-carbon metabolism. These pathways also affect the supply of NADPH. NADPH is distributed differently in the cytoplasm and mitochondria, and this partitioning indicates that it has different effects in different locations and is also important for local metabolic activities.

NADPH plays a central role in both anabolic and antioxidant activities. It provides reducing power to help synthesize important molecules, and it also helps scavenge reactive oxygen species (ROS), thereby protecting cells from oxidative stress. Cells need to maintain a balance between the production and use of NADPH. If this balance is disturbed, it may lead to diseases such as metabolic disorders, neurodegeneration, and cancer. In recent years, scientists have begun to study the dynamic changes of NADPH in cells more deeply through new technologies such as gene sensors and fluorescence imaging. These new discoveries have given us a better understanding of its regulatory mechanisms. In the future, regulating NADPH levels may become a new treatment method to enhance the ability of cells to resist stress or help treat diseases related to oxidative damage and metabolic imbalance.

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Conflict of Interest Disclosure

The authors affirm that this research was conducted without any commercial or financial relationships that could be construed as a potential conflict of interest.

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