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Review Article

Recent Advances in Mitochondrial Pyruvate Carrier Inhibitors

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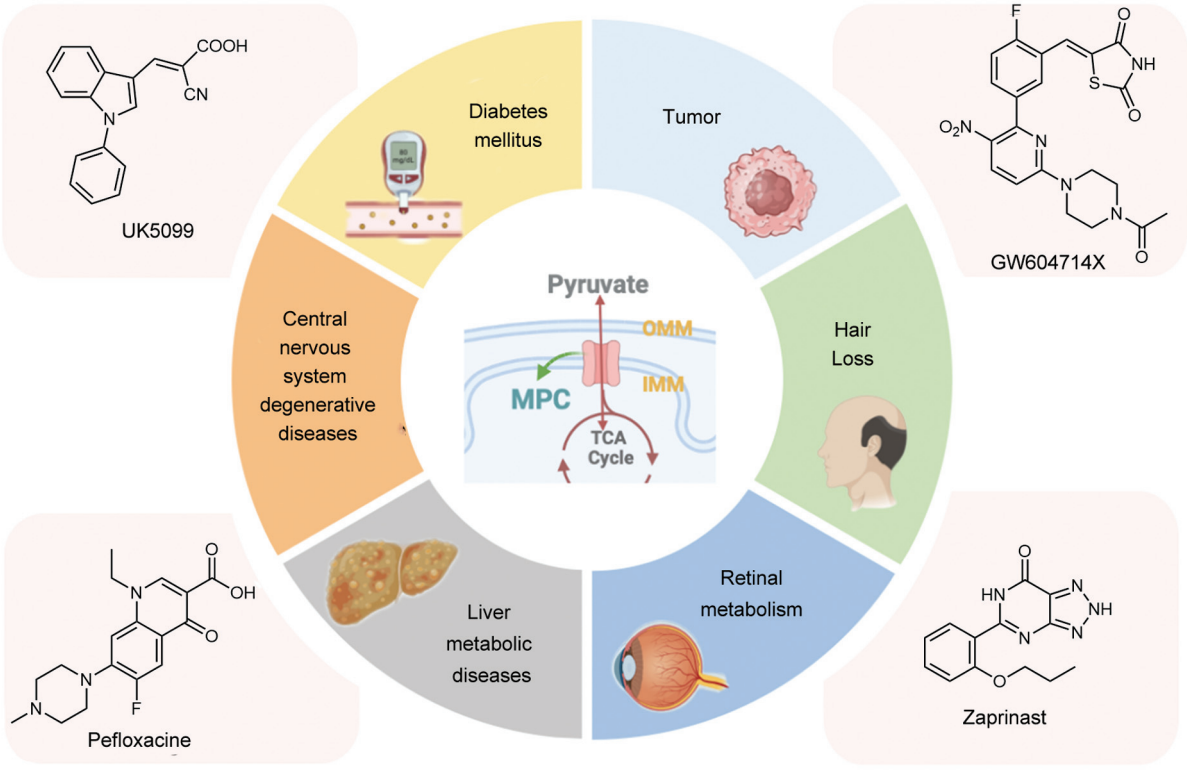

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Abstract

The mitochondrial pyruvate carrier (MPC) exists in the mitochondria inner membrane which transports pyruvate to the mitochondrial matrix. Evidence shows that MPC is the breakthrough point to study the regulation of basic energy metabolism, the dysfunction of which may lead to metabolic disturbance. Due to its important metabolic function, MPC has been considered a potential therapeutic target for diabetes, alopecia, cancers, neurodegenerative diseases, and liver metabolic diseases. However, MPC' protein crystal structure is still not clear as the proteins involved were only identified 10 years ago, making it difficult to carry out rational drug design based on receptor structure. In this review, we summarize the latest applications of MPC in different diseases and discuss the recent advances in pharmacochemical strategies of small-molecule inhibitors of MPC, hoping to promote the development of specific MPC inhibitors.

Keywords

- metabolism
- mitochondrial pyruvate carrier
- MPC inhibitors

Introduction

In eukaryotic cells, mitochondria are endosymbiotic organelles that are involved in a variety of cellular processes, including energy consumption, biosynthesis, signal transduction, and programmed cell death.[1] Significantly, they serve as the primary locations for the creation of adenosine triphosphate (ATP), the universal free energy carrier of all living things, including all five respiratory chain complexes and all tricarboxylic acid cycle (TCA) enzymes. Metabolite exchange between the cytoplasm and mitochondrial matrix is necessary to carry out these metabolic processes that are restricted to mitochondrial chambers and preserve intracellular homeostasis. The voltage-dependent anion channel allows tiny molecules to pass through the outer mitochondrial membrane. However, the inner mitochondrial membrane (IMM) is highly impermeable to molecules and ions and must rely on specific transporters and channels to connect the metabolism of the cytoplasm and mitochondria. The mitochondrial carrier family members perform the majority of the transport steps.[2] Other transporter families include mitochondrial pyruvate carrier (MPC).[3] MPC is a protein complex that exists in the mitochondrial inner membrane and is responsible for transporting pyruvate from mitochondria to the mitochondrial matrix in which pyruvate converts to acetyl-coenzyme A (acetyl-CoA). Acetyl-CoA enters the TCA cycle where it is further oxidized. Alternatively, pyruvate in the mitochondria can also participate in gluconeogenesis through the carboxylation of pyruvate carboxylase to produce oxaloacetic acid to supplement the TCA cycle. In addition to being transported to mitochondria as mentioned above, pyruvate can also be reduced to lactic acid by lactate dehydrogenase (LDH) in the cytoplasm.

MPC was first proposed in the 1970s[4] and was initially known as Brp44L (brain protein 44-like) and Brp44 (brain protein 44). It was identified in yeast in 2003 and further in mammals in 2012.[3,5,6] MPC is a relatively small heterooligomer consisting of two subunits, MPC1 and MPC2 (Brp44L and Brp44) of 12 and 14 KDa, respectively, in mammals.[7] MPC regulates energy metabolism by regulating the flux of pyruvate into the mitochondrial matrix. MPC is closely related to the occurrence and development of metabolic diseases. Currently, MPC has become a potential target for diabetes, alopecia, cancers, neurodegenerative diseases, liver metabolic diseases, etc. In recent years, more and more studies have focused on the structure, function, and inhibitory activity of MPC. Despite all this, the development of highly selective, nontoxic, and effective MPC inhibitors is still a huge challenge.

In this review, we summarized the correlation between MPC and different diseases, and suggest the pharmacochemical strategies for the discovery and development of small-molecule MPC inhibitors, hoping that this perspective provides ideas for the subsequent development of MPC inhibitors in these promising fields.

MPC and Its Involvement in Diseases

MPC Mechanism

Mitochondria provide energy by converting sugars, fats, and proteins into ATP through the TCA cycle coupled oxidative phosphorylation (OXPHOS). Pyruvate is located at an important crossroad of intermediate metabolism involved in the TCA cycle, oxidative metabolism, gluconeogenesis, lipid de novo synthesis, and cholesterol synthesis.[3] Pyruvate is a product of glycolysis in the cytoplasm (►**Fig. 1**), and is transported by the MPC in the IMM to the mitochondrial matrix to participate in the metabolic process.[8]

In 1971, Papa et al confirmed that pyruvate crossed the mitochondrial membrane through a specific transporter and put forward the concept of MPC for the first time.[4] MPC was named Brp44L and Brp44 based on the location of protein expression found at that time.[4,9] MPC, a pyruvate transporter located on the IIMM, was first identified in yeast in 2003.[5] In 2012, Bricker et al found that the MPC is also present in mammals and composed of MPC1 and MPC2 proteins.[3] Silencing of *MPC1* or *MPC2* in mammalian cells impairs the oxidation of pyruvate.[3] In the same year, Herzig et al reported that MPC in yeast consists of Mpc1 and Mpc2 or Mpc3.[6] The MPC, which consists of MPC1 and MPC2 in mammals, is a hetero-dimer.[7] In *Saccharomyces cerevisiae*, the expression of the three proteins (Mpc1, Mpc2, and Mpc3) depends on the

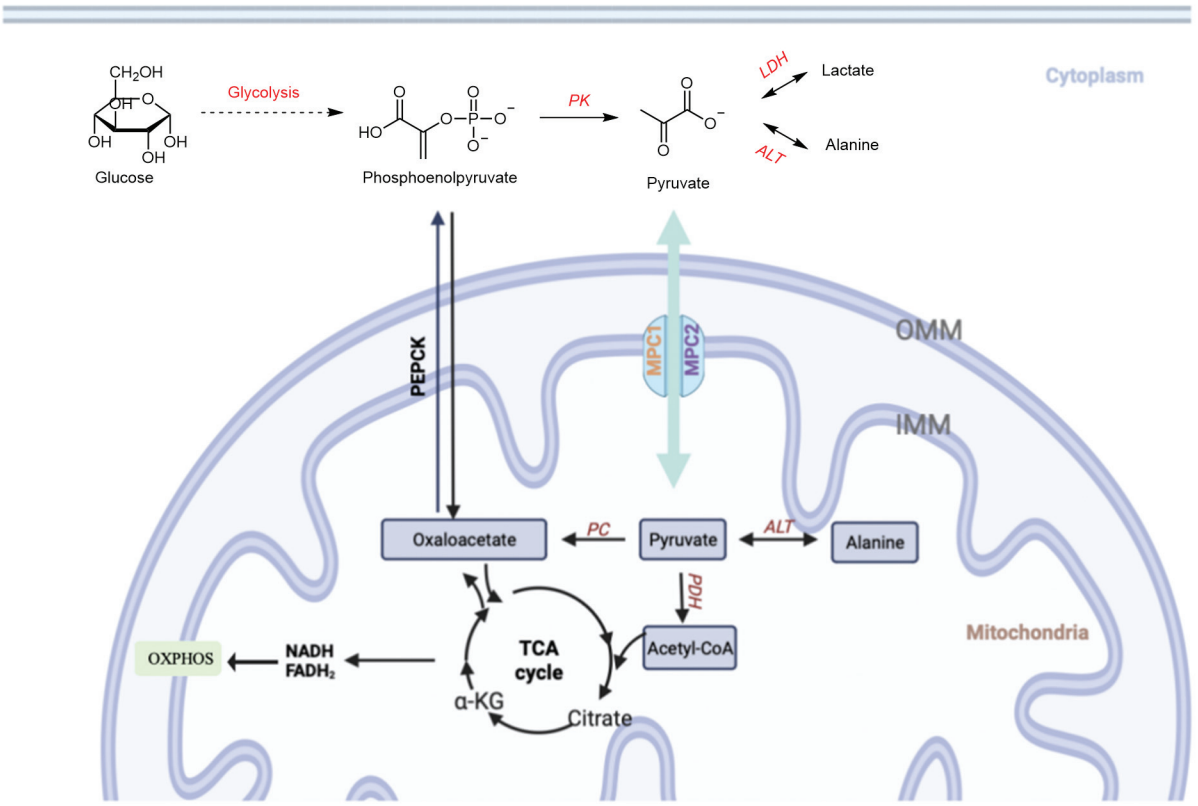


Fig. 1 The mechanism of MPC action: mitochondria provide energy by converting sugars, fats, and proteins into ATP through TCA-coupled oxidative phosphorylation. Pyruvate is located at the important crossroads of intermediate metabolism involving gluconeogenesis, oxidative metabolism, lipid de novo synthesis, cholesterol synthesis, and the TCA cycle. The image was created with BioRender.com. ATP, adenosine triphosphate; TCA, tricarboxylic acid cycle; IMM, inner mitochondrial membrane; MPC, mitochondrial pyruvate carrier; OMM, outer mitochondrial membrane.

carbon source. These proteins combine to form the MPCFERM and MPCOX complexes, which are named for the Mpc1/Mpc2 complex formed under fermentative conditions and the Mpc1/Mpc3 complex formed under respiratory conditions, respectively.[1] Tavoulari et al found that MPC is a heterodimer rather than a multimeric complex. The yeast Mpc proteins can also form inactive homo-dimers when they are not involved in hetero-dimers.[7]

Deletion or knock-down of any MPC subunit results in heterodimer degradation, so knockout of *Mpc1* or *Mpc2* is often considered a dual deficiency of MPC protein and activity. Abnormal expression of either subunit results in MPC dysfunction. MPC plays a role in maintaining the balance between glycolysis and OXPHOS, and MPC abnormalities may be accompanied by metabolic disturbances.[3,6,10] Germline deletion of the *Mpc* gene was found to cause early embryo death in mice,[11–13] and mutations in the *MPC1* and *MPC2* in the population were also found to cause significant growth retardation and death in utero or childhood.[14,15] Lactic acidosis, hypotonia, malformation, and encephalopathy are the most common symptoms of MPC mutations with loss of function, and these patients are often in critical condition.[14,15] Conditional loss of MPC in the heart[10] and central nervous system (CNS)[16] of mice leads to progressive cardiomyopathy and sensitivity to epileptic seizures, respectively.

Diabetes Mellitus

Gluconeogenesis is essential to maintain appropriate blood sugar levels while fasting, and elevated gluconeogenesis in patients with type 2 diabetes mellitus (T2DM) leads to chronic hyperglycemia. Gray et al have shown that MPCs including MPC1 and MPC2 proteins are required to efficiently control hepatic gluconeogenesis (►**Fig. 2**).[17] Liver mitochondrial pyruvate uptake and pyruvate-driven respiration depend on MPC. The *Mpc1* liver-specific knockout mice had overall changes in metabolism but were able to maintain fasting euglycemia. Pyruvate-driven hepatic gluconeogenesis is dependent on MPC. *Mpc1* deletion impairs the flux of the pyruvate-driven TCA cycle and reduces stable fasting liver glutamine concentration while enhancing indicators of urea cycle activity. Dietary obesity increases MPC expression and activity in the liver. Acute *Mpc1* deletion after dietary obesity reduces hyperglycemia and improves glucose tolerance. Constitutive loss of liver MPC activity attenuates the development of hyperglycemia and glucose intolerance associated with obesity induced by a high-fat diet.

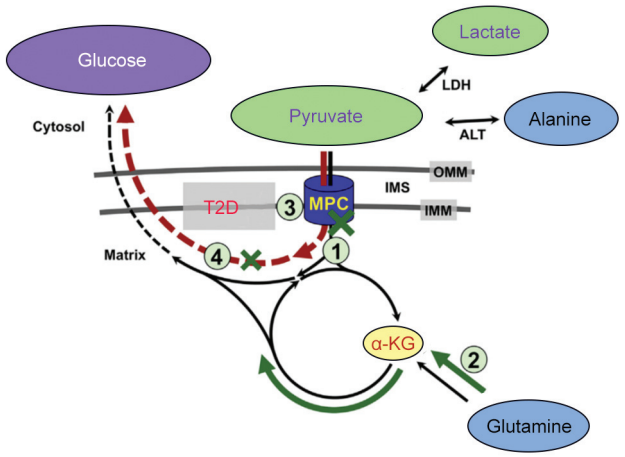


Fig. 2 The role that MPC plays in type 2 diabetes. MPC, mitochondrial pyruvate carrier. (Reprinted with permission from Gray LR, Sultana MR, Sultana MR, Rauckhorst AJ, et al. Hepatic mitochondrial pyruvate carrier 1 is required for efficient regulation of gluconeogenesis and whole-body glucose homeostasis. Cell Metab 2015;22(04):669–681.[17])

McCommis et al used embryonic stem cell technology to obtain mice with liver-specific deletion of MPC2 (LS-*Mpc2*$^{-/-}$).[18] They found that the ability of hepatocytes to convert tagged pyruvate into TCA cycle intermediates and glucose was damaged, but not eliminated, by MPC2 loss. Although pyruvate has defects in gluconeogenesis, LS-*Mpc2*$^{-/-}$mice have a normal appearance, no obvious liver pathological evidence, and only show slight hypoglycemia in prolonged fasting studies. Additionally, it is proved that the pyruvate–alanine cycle in LS-*Mpc2*$^{-/-}$mice may constitute an alternative pathway of pyruvate/lactate gluconeogenesis by bypassing the MPC complex. Therefore, treating diabetes and other related metabolic illnesses with selective targeting of MPC complex inhibitors is a practical approach.

Hair Loss

Hair follicle stem cells (HFSCs) are normally dormant, but they are rapidly activated during the new hair cycle due to the influence of various cytokines (BMP, WNT, etc.).[19] When HFSCs are inhibited or inactivated, hair loss results. Numerous internal and external factors control the static state of HFSCs, although the *in vivo* metabolic pathways of the hair follicle are unclear.

Lactic acid generation is crucial for the activation of HFSCs. LDH is an enzyme that catalyzes the reduction of pyruvate to lactic acid. By knocking out the *Mpc* gene or inhibiting MPC activity with an MPC inhibitor (UK-5099), LDH activity is increased in HFSCs, which promotes lactic acid production and hair growth.[20] At the same time, low expression of MPC1 encouraged LDH to convert pyruvate into lactic acid.[21]

Tumor

Glycolysis is the process of converting glucose into lactic acid, which produces two molecules of ATP per molecule of glucose. OXPHOS is the process by which aerobic cells convert glucose to pyruvate. Pyruvate is then transported into mitochondria where it is oxidized to make 36 molecules of ATP. Normal differentiated cells rely primarily on mitochondrial OXPHOS to provide energy for cells and partly through glycolysis[1]; however, the majority of tumor cells rely on aerobic glycolysis, which converts pyruvate metabolism from mitochondrial OXPHOS to cytoplasmic glycolysis to support tumor proliferation. This phenomenon is known as the "Warburg effect."[22] Whether the cells use glycolysis or OXPHOS to produce energy depends on whether pyruvate can enter the mitochondria.

It has been shown that MPC1 and MPC2 are downexpressed in cancer, and this low expression is associated with low survival in several cancers, including lung, kidney, and colon cancer.[21] All tumors found had low levels of *MPC1*, except for leukemia and lymphoma, which are hematological malignancies. In some malignancies, *MPC2* is overexpressed, while in others, it is underexpressed.[23] Data from colon adenocarcinoma are particularly striking, with the abundance of *MPC1* and *MPC2* mRNAs in tumors being much lower than that in normal tissues. MPC suppression promotes the "Warburg effect" and maintains the stemness of colon cancer cells.[21] Regulation of the MPC complex is the key to tumor cell proliferation.

Furthermore, five distinct cancer types were identified based on notable alterations in MPC1 expression: lung adenocarcinoma (LUAD), pancreatic adenocarcinoma (PAAD), prostate adenocarcinoma (PRAD), kidney renal clear cell carcinoma (KIRC), and breast invasive carcinoma (BRCA). Low MPC1 expression is substantially correlated with shorter overall survival in BRCA, while high MPC1 expression is closely linked to a favorable prognosis in KIRC, LUAD, PAAD, and PRAD. MPC1 expression is significantly positively and negatively correlated with immune cell infiltration in thymoma and thyroid cancer.[24]

The potential metabolic effect of MPC1 on cancer is reflected in reduced pyruvate transport capacity[25]; impairment of pyruvate-driven OXPHOS; and increased lactate production, glucose consumption, and glycolysis capacity,[26] as well as the underlying mechanism.[24] These effects promote tumor invasion, migration, and progression. MPC1 is a new cancer biomarker with the potential to be an effective target for cancer detection and management. Research directed at reducing cancer progression is currently underway.

Central Nervous System Degenerative Diseases

CNS degenerative diseases are a kind of disease characterized by the gradual loss of the structure and function of the CNS.[27] Typical degenerative diseases of the CNS include Alzheimer's disease (AD), Parkinson's disease (PD), Huntington's chorea, amyotrophic lateral sclerosis, etc.

Parker found that the mitochondrial genome may play a role in the pathogenesis of AD, and the pathophysiology of CNS degenerative disorders is largely dependent on mitochondrial damage.[28] MPC is widely distributed in the human body including the CNS, such as the thalamus, hippocampus, cerebral cortex, and dopaminergic neurons.[29] In experimental models of degenerative diseases, the pyruvate transport

restriction by inhibiting MPC attenuates inflammatory response and enhanced neuroprotection.[30]

Thiazolidinediones (TZDs), including pyrrolidone and rosiglitazone, are commonly used antidiabetic drugs and are also a class of MPC inhibitors.[31] Clinical experiments show that pyrrolidone and rosiglitazone can improve cognitive performance in patients with AD.[32] Over 14 years, diabetics taking TZDs had a 29% lower risk of developing PD.[33] However, MPC plays an important role in mitochondrial energy supply. If it is excessively inhibited, it may cause damage to cells, including nerve cells.[34] Therefore, the regulation of MPC targets can provide a new therapeutic direction for neurological dysfunction caused by degenerative diseases of the CNS.

Other Diseases

Liver diseases associated with metabolic syndrome have become the most common liver metabolic diseases, including nonalcoholic steatohepatitis (NASH), but its specific mechanism of action is still unclear and there is currently no Food and Drug Administration-approved drug for it on the market.[35] Hepatic stellate cells (HSCs) are nonparenchymal in the liver and are the primary cause of fibrosis in various liver diseases. Ferguson et al found that MPC inhibitor 7-ACC2 inhibited markers of HSC activation and reduced fibrosis in NASH mice models.[36] McCommis et al found that TZDs, including MSDC-0602, bind to MPC2 in mitochondria. In addition to preventing and reversing liver fibrosis, MSDC-0602 reduces the expression of HSC activation markers in mouse liver. Knockout of *Mpc2* also affects the secretion of exosomes from hepatocytes and inhibits the activation of HSC.[37] Therefore, targeting MPC2 provides a fresh approach to the therapy of liver metabolic diseases.

Aerobic glycolysis dominates retinal metabolism (Warburg effect), and mitochondria oxidize a tiny portion of the pyruvate generated by glycolysis.[38] Localized MPC1 deletion in the retina leads to progressive retina degeneration and reduced visual function of the rod and cone cells. In the retina, mitochondrial pyruvate transport is essential for preserving photoreceptor integrity as well as controlling substrate consumption and neurotransmitter production.[39]

Research Advances of MPC Inhibitors

Halestrap and Denton discovered MPC inhibitors in 1974 and found that α-cyano-4 hydroxycinnamate (CHC) binds to specific proteins on the IMM and specifically inhibits pyruvate transport without inhibiting enzymes involved in pyruvate metabolism.[40] CHC esters and their analogs are exceptionally effective in inhibiting pyruvate transport compared to other carboxyl carrier inhibitors, among which UK5099 is a more efficient noncompetitive inhibitor.[9] Divakaruni et al reported that TZDs, commonly used as insulin sensitizers, acutely and specifically inhibit MPC activity.[41] In recent years, quinolone antibiotics, coumarin derivatives, and other compounds acting on MPC have been discovered, which has promoted the research progress of pharmacology and medicinal chemistry.

Pharmacological Mechanism of MPC Inhibitors

CHC and its analogs react reversely with mercaptoethanol and cysteine to generate additional products and react with essential mercaptan groups on the pyruvate carrier.[42] It was initially thought that the binding of CHC or UK5099 to MPC was noncompetitive and that this inhibition was reversible. The oxygen uptake in mitochondria was gradually enhanced from virtual total inhibition to almost the same level as control mitochondria that had been preincubated without inhibitors by first incubating the mitochondria with inhibitors and then diluting the mitochondria into an oxygen electrode incubation mixture.[9] This binding could occur because the inhibitor forms a covalent bond with the carrier, causing the product to hydrolyze slowly, or it could occur because the inhibitor is noncovalent and slowly attaches to specific places on the carrier protein, rather than the active center, distorting the carrier and rendering it ineffective. UK5099 has been reported to bind covalently and irreversibly to the mercaptan group of MPC1.[43] Unlike UK5099, the action of TZDs inhibitors is thought to be reversible, with the rate of pyruvate oxidation returning to that of the blank group after washing permeation.[41]

Recently, Yamashita et al used a YY4-yne probe to overturn the previous hypothesis that UK-5099 binds to a cysteine on MPC1, indicating that it may bind to cysteine-54 of MPC2.[44] However, it is still possible for UK-5099 to inhibit MPC by binding to MPC1 or at the interface between MPC1 and MPC2.[43] It is difficult to exactly characterize the role and binding mode of MPC transporters in mitochondria because MPC genes and proteins have not been purified.

Detection Methods Targeting MPC

MPC function was first investigated in the 1970s using mitochondria isolated from mammals, and the discovery of molecular properties of MPC and its potential as a therapeutic target sparked further interest in developing novel functional tests.[45] Currently, little is known about the composition and workings of MPC. Thus, in terms of MPC physiological function pathways, the primary assays include lactate release assay; bioluminescence resonance energy transfer (BRET) ligand-binding assay; substrate transport and transport inhibition in isolated mitochondria; thermostability shift assays to evaluate binding; substrate transport and transport inhibition in proteoliposomes prepared with purified protein, among other techniques. Compound discovery and optimization techniques are applied. In this article, we summarized the fundamentals, advantages, and disadvantages of the assays commonly used to detect and evaluate MPC inhibitors (►**Table 1**).

Isolated mammalian mitochondria were cultured in a solution containing radio-labeled pyruvate, which was brought into the matrix by the MPC. The mitochondria were quickly centrifuged to terminate the process, or separated from the uptake buffer by membrane filtration.[46,47] Current methods for tracking pyruvate inflow into mitochondria can only indirectly measure MPC activity. We named RESPYR (for Reporter Sensitive to PYRuvate) after Compan et al' BRET-based genetically encoded biosensor.[48]

Table 1 An overview and comparison of the assays used to measure MPC

Detection method	Introduction to the methodology	Advantages	Disadvantages	Ref.
Substrate transport and transport inhibition in isolated mitochondria	Mitochondria isolated from mammals were incubated in a buffer containing radiolabeled pyruvate, which was transported into the matrix by MPC, and the reaction was subsequently stopped by centrifugation or membrane filtration.	Detection in natural membrane environments.	Intact mitochondria are not easy to isolate; specificity is difficult to determine.	46,47
BRET ligand-binding assay	MPC1 and MPC2 were modified by C-terminal fusion to the donor group RLuc8 (a variant of Renilla luciferase) or the recipient group Venus (a variant of yellow fluorescent protein), and MPC activity was monitored in real-time by monitoring changes in BRET.	MPC activity can be monitored in real time and the measured changes in MPC activity are fully reversible.	Ectopic expression of the two labeled subunits dependent on the vector may result in excessive or uneven levels of subunit expression or conformational changes that impair the pyruvate transport base.	48
Thermostability shift assays to evaluate binding	Proteins begin to unfold as they undergo temperature ramps, and in combination with detectable changes, the apparent melting temperature of the protein population can be calculated.	High throughput, direct assessment of binding of purified MPCs; highly sensitive, easy to implement, reproducible; validation of folded carrier proteins.	Higher cost and inability to directly determine binding parameters.	7,49–52
Lactate release assay	Loss of function of MPC1 drives lactate production by enhancing the conversion of pyruvate to lactate by LDH.	Not reported	Not reported	20,21,53
Substrate transport and transport inhibition in proteoliposomes prepared with purified protein	Preparation of liposomes using phosphatidylcholine and cardiolipin relies on radial substrate transport, rapid filtration to separate liposomes, and a scintillation counter for radioactivity counting.	It is easier to apply and control chemical gradients, and any effector can be tested directly on MPC proteins; reliable quantitative data can be collected for mechanistic studies or inhibitor screening.	The activity is not comparable to that measured in natural membranes; not a high-throughput solution for compound screening.	7,49
Pyruvate transport and inhibition in the *Lactococcus lactis*	Expression of mouse MPC1 and MPC2 alone or in combination in *Lactococcus lactis* and compared to the empty vector control.	Not reported	Not reported	6
Chemical proteomics probes for MPC	Alkyne-modified Aca, YY4-yne, can be used as a versatile cell-targeted splicing probe for MPC2 in click chemistry-activated Western blotting or whole-mass spectrometry-based proteomics experiments.	Not reported	Not reported	44

Abbreviations: BRET, bioluminescence resonance energy transfer; LDH, lactate dehydrogenase; MPC, mitochondrial pyruvate carrier.

MPC1 and MPC2 are altered by C-terminal fusion to either acceptor group Venus (a variant of yellow fluorescent protein) or donor group RLuc8 (a version of Renilla luciferase). Real-time monitoring of MPC activity with high temporal precision can be achieved by observing changes in BRET in different cell types and circumstances (►**Fig. 3**).[48] The method is based on the assumption that during ligand binding, conformational changes occur that alter the closeness between the termini, which results in an energy transfer between MPC1 and MPC2.[45]

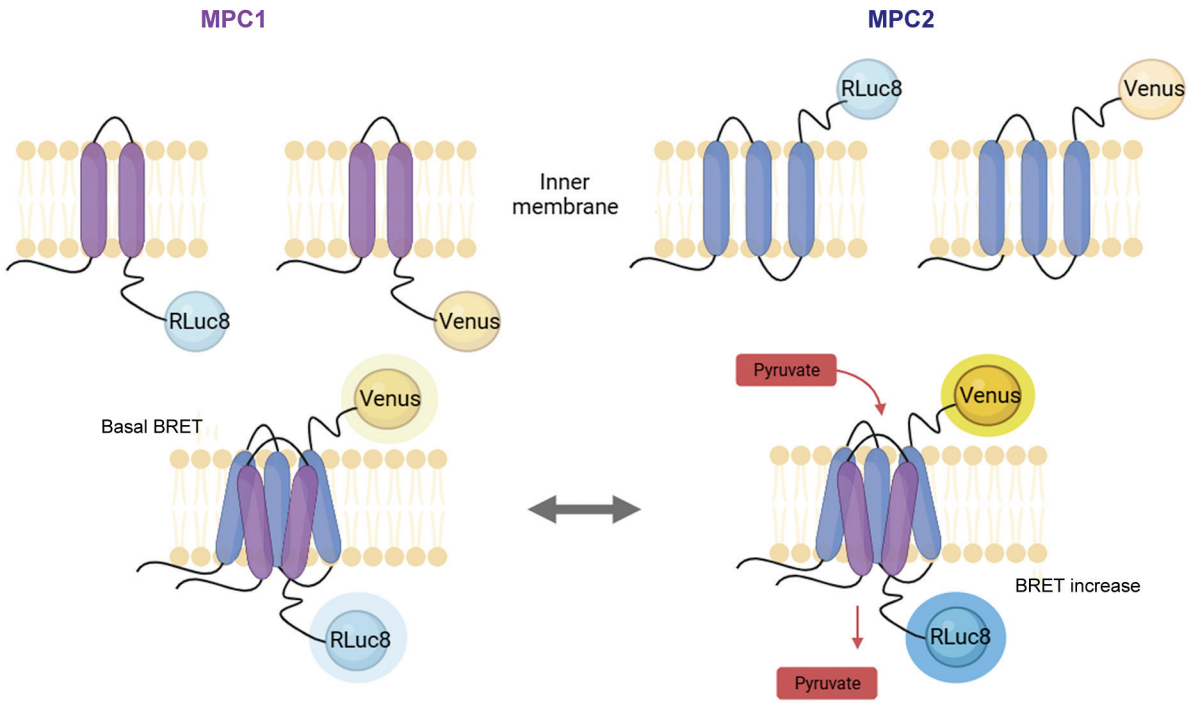


Fig. 3 Mechanisms that increase BRET signaling. The image is created with BioRender.com. BRET, bioluminescence resonance energy transfer.

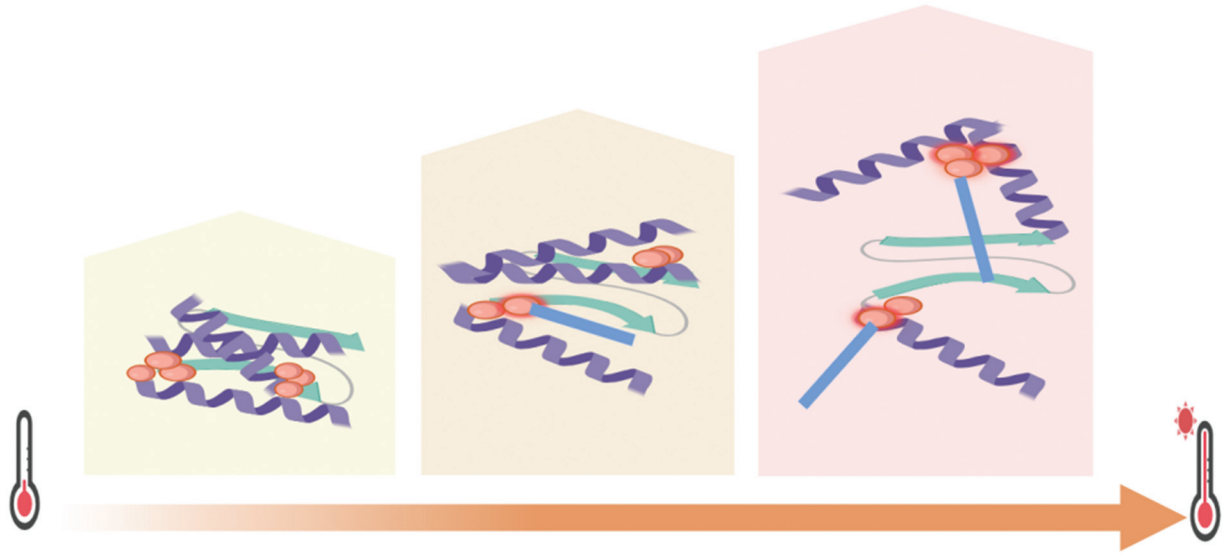


Fig. 4 When the temperature is steadily increased (25–90°C), protein molecules in the population unfold, and buried cysteine residues are exposed to the solvent and accessible to the thiol-specific probe CPM (blue stick depiction) and become fluorescent upon reaction. The image is created with BioRender.com. CPM, 7-diethylamino-3-(4-maleimidophenyl)-4-methylcoumarin.

Two thermal stability transfer assays can be used to monitor ligand binding to purified MPC (►**Fig. 4**).[49] The first thermal stability shift assay used the reaction of endogenous cysteine with 7-diethylamino-3-(4-maleimidophenyl)-4-methylcoumarin (CPM) to measure the change in thermal stability of the proteins upon ligand binding.[50] The second technique, known as dye-free nano differential scanning fluorimetry (nano-DSF), tracks how the surroundings of the tryptophan and tyrosine residues change during unfolding, causing changes in fluorescence.[7] In addition, it can detect MPC function by promoting lactate release assays. According to Lowry's group, pharmacological inhibition of MPC by UK5099 or genetic deletion of the MPC can stimulate LDH activity, which in turn increases LDH activity in HFSCs.[20] When MPC is knocked down or pharmacologically blocked, pyruvate in the cytoplasm does not enter the mitochondria,

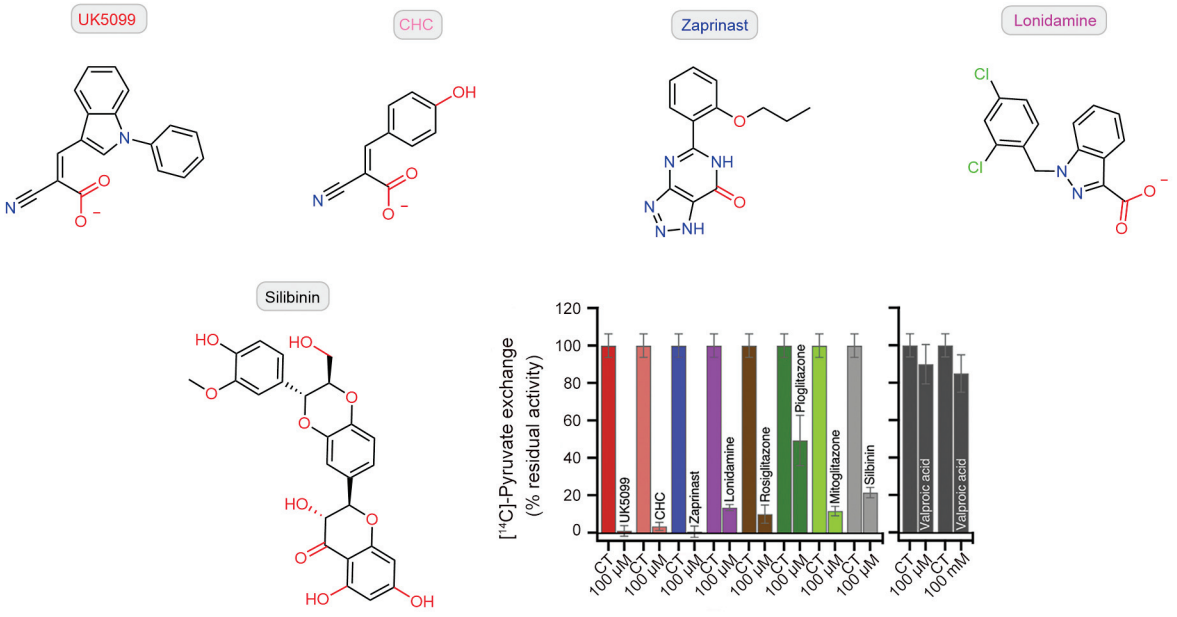


Fig. 5 Tavoulari et al revealed the inhibition of pyruvate transport of the drugs listed at a concentration of 100 mmol/L on MPC1L/MPC2 proteoliposomes, and the residual transport activity was compared to that in their absence (CT). (Reprinted with permission from Tavoulari S, Schirris TJJ, Mavridou V, et al. Key features of inhibitor binding to the human mitochondrial pyruvate carrier hetero-dimer. Mol Metab 2022;60:101469.[49])

as seen by *in situ* LDH. Instead, it is converted to lactate, another metabolite, via LDH, which increases LDH activity and activates HFSCs.[53]

Reconstitution of pure proteins into liposomes is another useful technique for studying transport. In this method, the transport of pyruvate in phosphatidylcholine (PC)/cardiolipin (TOCL) liposomes was investigated by human and yeast heterodimers.[45] Tavoulari et al used this method to test the ability of different classes of MPC inhibitors to inhibit MPC1/MPC2 pyruvate transport at a concentration of 100 mmol/L and compared their residual transport activity to that in the absence. At this concentration, pyruvate transport was completely inhibited by UK5099, CHC, and zaprinast, and to a lesser extent by lonidamine, TZDs, and silibinin (►**Fig. 5**).[49]

Other techniques include monitoring pyruvate uptake in comparison to empty vector control and expressing mouse MPC1 (mMPC1) and mMPC2 alone or in combination in *Lactococcus lactis*.[6] A chemical proteomics probe using MPC confirmed that α-chloroacetamide (αCA) binds strongly to cysteine-54 (C54) in subunit 2 of MPC (MPC2). This led to the generation of the alkyne-modified αCA, YY4-yne, which served as a versatile cell-target splicing probe for MPC2 in click chemistry-activated Western blotting or whole-mass spectrometry-based proteomics experiments (►**Fig. 6**). The results show that UK-5099 binds to C54 of MPC2 to block the MPC complex covalently and reversibly. The YY4-yne probe can be used to measure this inhibition in cells.[44]

Research Progress on Classification and Pharmacochemistry of MPC Inhibitors

As mentioned earlier, the structure of the MPC protein is unknown, but a variety of activity assays are available. Small-molecule MPC inhibitors of multiple structural types have been reported to date. Herein, the discovery and optimization process of these small-molecule inhibitors were reviewed.

Cyano-cinnamates and Their Analogues

The first molecules identified as MPC inhibitors were α-cyano-4-hydroxycinnamate (CHC) and its derivatives, which inhibit pyruvate transport and prevent its oxidation in intact mitochondria. ►**Table 2** summarizes the inhibitory activity of CHC analogs in pyruvate transport in rat liver and heart mitochondria, where the important structural features may

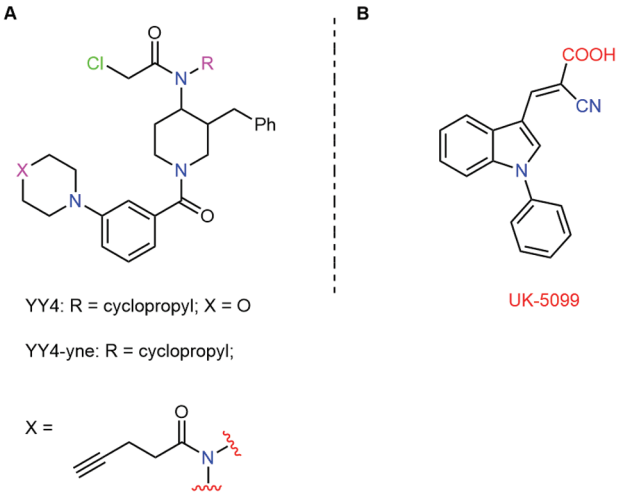
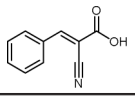
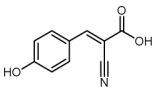
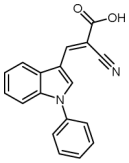
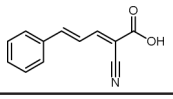
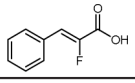
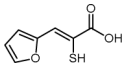
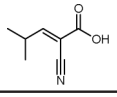


Fig. 6 Yamashita et al demonstrated that UK-5099 may bind to C54 of MPC2 using the YY4-yne probe.[44] The image showed the chemical structures of the compounds tested in the study: (**A**) αCAs YY4 and corresponding alkyne probe YY4-yne, as well as (**B**) MPC inhibitor UK-5099.

Table 2 The inhibitory activity of CHC analogs on pyruvate transport in rat heart mitochondria

Name	Structure	MPC IC_{50} (μmol/L)
α-Cyanocinnamate		0.2
CHC		1.5
UK 5099		0.05
α-Cyano-5-phenyl-2, 4-pentadienoate		0.2
α-fluorocinnamate		200
α-Thio-2-furanpyruvate		500
α-Cyano-4-methyl-2-pentanoate		>1,000

Abbreviations: CHC, α-cyano-4-hydroxycinnamate; UK 5099, α-cyano-β-(1-phenylindol-3-yl) acrylate.
Source: Reproduced with permission from Halestrap AP. The mitochondrial pyruvate carrier. Kinetics and specificity for substrates and inhibitors. Biochem J 1975;148(01):85-96.[9]

be the nitrile group and the aromatic side chain. Among them, UK 5099 [α-cyano-β-(1-phenylindol-3-yl)acrylate] is the most potent.

As shown in ►**Table 2**, the inhibitory activities of α-cyanocinnamate (IC_{50} = 0.2 μmol/L), α-cyano-5-phenyl-2,4-pentadienoate (IC_{50} = 0.2 μmol/L), and UK 5099 (IC_{50} = 0.05 μmol/L) were stronger than that of CHC (IC_{50} = 1.5 μmol/L). The inhibitory activity of α-fluorocinnamate (IC_{50} = 200 μmol/L) is 1,000-fold lower compared with that of α-cyanocinnamate, indicating the importance of the nitrile group as a pharmacodynamic functional group; moreover, the IC_{50} value of α-cyanocinnamate and α-cyano-5-phenyl-2,4-pentadienoate were the same, which suggested that addition of two carbons to the side chain does not have much effect on the inhibitory activity of the compounds.

When the benzene ring of α-cyanocinnamate was replaced with an alkyl group, α-cyano-4-methyl-2-pentanoate was obtained, which did not exhibit any inhibitory effect, even at a high dose of 1 mmol/L. When the 4-position of the benzene ring of α-cyanocinnamate was replaced by a hydroxyl group, CHC was obtained with its activity being reduced greatly. Given above, the benzene ring and the hydrophobic properties of the derivates are important in inhibiting MPC activity.[9]

Hydrophobic aromatic moieties may lock the active α-cyanocinnamate group to the necessary inhibition positions on the support. In addition, it has been reported that hydrophobic pockets may exist on the surface of the carrier, and that inhibitory activity may increase with increasing hydrophobicity. The inhibitors can reach the site by binding to the outer surface of the carrier, undergoing a conformational change, and then transferring to hydrophobic matrices facing the binding pockets.[43]

In 2021, Liu et al reported MPC inhibitors for the treatment of alopecia.[53] Using UK5099 as a lead compound, the N1 position, substitutes on the indole core, various aromatic and heteroaromatic core structures, and the variable Michael acceptors were modified. The inhibition of MPC was validated by monitoring the rate of lactic acid production in cells

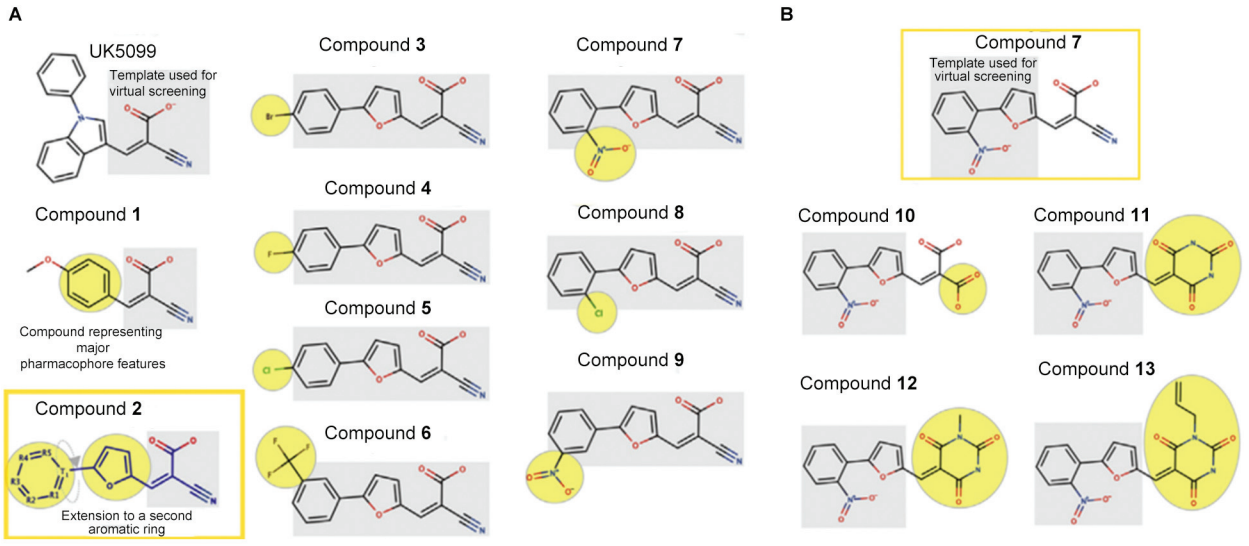


Fig. 7 (A) and (B) were produced by screening compounds with templates of UK5099 and compound **7**, respectively. The gray box indicates the virtual screening template. Yellow indicates the newly added or modified chemical groups. (Reprinted with permission from Tavoulari S, Schirris TJJ, et al. Key features of inhibitor binding to the human mitochondrial pyruvate carrier hetero-dimer. Mol Metab 2022;60:101469.[49])

and a more thorough understanding of the structure–activity relationship (SAR) of MPC inhibitors was obtained.[53]

The effect of substituents at the N1 position was examined. When the phenyl group was replaced with H, JXL002 was obtained with little inhibitory activity, suggesting the importance of the phenyl group for the activity of UK5099. Lengthening the carbon chain between the indole ring and the benzene ring (JXL079) or adding a substituent, such as methoxy to the benzene ring (JXL006), will decrease the activity. In addition, the 3,5-bis(trifluoromethyl)benzyl group was the best moiety at the N1-position of the indole core in this section (JXL020).

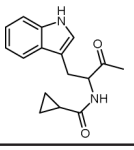
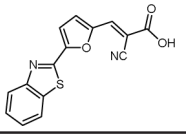
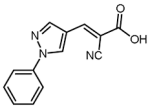
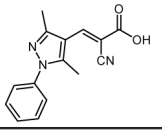
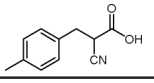
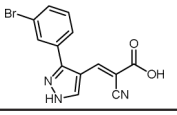
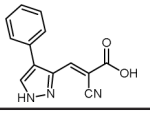
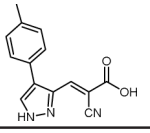
The effect of substituents on the indole ring was further investigated. It was found that large substituents are harmful to inhibit MPC activity, probably due to the spacing effect of the large substituents, whereas small substituents at the 4-position are beneficial. However, the introduction of highly polar groups, such as carboxyl groups, is not a good idea because highly polar groups may have difficulty penetrating the cell membrane.

Aromatic rings and heterocyclic aromatic compounds other than the indole ring were not prominent in promoting lactic acid production, indicating that the indole ring is important for maintaining the activity. The effect of the Michael receptor, which can partly change the lipophilicity and electrophilicity of molecules, was investigated. The data showed that only the ethyl ester was as potent as the carboxyl group, while the methyl ester and the *tert*-butyl ester analogs showed a greater decrease in potency. The amide analogs greatly increase the ability to produce lactic acid but cause substantial cell death. Adjusting the Michael receptor would therefore significantly affect the biological activity.[53]

Tavoulari et al compared all different types of MPC inhibitors and found that three closely arranged hydrogen bond acceptors and an aromatic ring are the common characteristics of all inhibitors and represent the minimum requirement for high efficiency. Having two hydrogen bond acceptors may be the minimum requirement for binding to MPC. In CHC analogs, hydrogen bond acceptors are represented by carboxyl and cyano groups. Hydrogen bond acceptors with similar functions to carboxyl and cyano groups are provided as requirements for embodied activity. All MPC inhibitors have a central aromatic ring moiety, which seems to be another requirement for MPC inhibition, second only to hydrogen bond acceptors. This is an additional feature that may increase the number of interactions between the inhibitor and MPC, thus improving affinity. Furthermore, the addition of nitro groups to the aromatic ring moiety may increase the inhibition efficacy (compound **7**) (►**Fig. 7**).[49]

In 2022, Hegazy et al developed a virtual screening prediction model for MPC inhibitors using the chemical and geometric properties of UK-5099.[54] The model consists of three parts, a negative charge corresponding to a carboxyl group, a hydrogen bond receptor corresponding to a cyano group, and an aromatic ring corresponding to indole ring. Using this approach, five new nonindole MPC inhibitors were found (►**Table 3**, BE1976, BE1978, BE1980, BE1984, and BE1985), four of which had IC_{50} values in the nanomolar range, and were up to seven times more powerful than the canonical inhibitor UK-5099. These new compounds exhibit characteristics of drugs and match Lipinski's five rules, with good aqueous solubility, oral bioavailability, and metabolic stability.[54] Among these compounds, BE1976 and BE1978 were the most potent inhibitors of MPC respiration. BE1976 is seven times more active than UK5099. Removal of the hydrogen bond receptor (cyano) eliminated the activity, demonstrating that the hydrogen bond receptor is essential. These compounds have novel structural features compared to the mother nucleus structure of common MPC inhibitors and offer fresh insight into the design of novel MPC inhibitors in the future.[54]

Table 3 Relative IC_{50} values of manufactured and discovered inhibitors of MPC from a reported study[54]

Name	Structure	MPC IC_{50} (μmol/L)
BE1975		Inactive
BE1976		0.033
BE1978		0.117
BE1980		0.162
BE1984		1.53
BE1985		0.638
BE2617		0.039
BE2623		0.731

Abbreviation: MPC, mitochondrial pyruvate carrier.

Thiazolidinediones

Insulin sensitizers and agonists of the nuclear receptor transcription factor peroxisome proliferation-activated receptor gamma (PPARγ) are TZDs, including pioglitazone and

rosiglitazone.[55] Many of the benefits of TZDs are mediated through this receptor. But in addition to interacting with transcriptional regulators, TZDs can affect metabolism through other molecular targets.[31,41,56] Pharmacological activity independent of PPARγ is associated with the ability of these drugs to interact with the MPC complex.[31,41] On the other hand, conditional deletion of any MPC protein in hepatocytes is safe against additional metabolic problems associated with a high-fat diet, including diabetes and liver damage.[17,18,37,57] This is consistent with the role of MPC-targeted TZDs in inhibiting pyruvate entry into the mitochondrial metabolic pathway in an inhibitory manner.[58]

A class of thiazolidinediones were found to be known as insulin sensitizers.[59] These substances, formerly referred to as insulin sensitizers,[59] are the best medications for stopping the progression of hyperglycemia into type 2 diabetes.[46] The reintroduction of pioglitazone for the treatment of neurological illnesses and some malignancies is the result of growing interest in the connection between human disease and disruption of glucose metabolism.[9,60,61] However, the greater therapeutic applicability of TZDs is limited, due to their substantial adverse effects, including volume enlargement, bone loss, increased obesity, and cardiovascular risk.[47,62] In the middle of the 1990s, only three drugs with this effect were licensed and marketed for the treatment of type 2 diabetes. Approximately 10 years later, it was discovered that these substances worked by directly activating PPAR.[59]

In addition, mitochondrial binding sites for TZDs were identified a few years earlier, and we refer to this complex as the mitochondrial target of mTOT TZDs.[63] Colca et al created a photocatalytic affinity probe based on the structure of pioglitazone. This approach provides a molecular tool that can covalently bind to it, once specifically bound to its site of action. Selective competition before probe photoactivation demonstrates specificity.[64] As seen with active TZD, the addition of UK-5099 to the experiment prevented the TZD probe from cross-linking to MPC2, demonstrating strong binding between the two classes of chemicals. This suggests that TZD which is sensitive to insulin may control the use of pyruvate.[31]

The first-generation TZDs, pioglitazone, and rosiglitazone, facilitate glucose/lipid uptake and storage in peripheral tissues (such as skeletal muscle, liver, and adipose tissue) by binding to and activating the transcription factor PPARγ, which in turn enhances insulin sensitivity. Although all TZDs have a comparable affinity for PPARγ, rosiglitazone is by far the most effective. In contrast, the newer generation of TZDs does not bind to or activate PPARγ. Studies have shown that various active TZDs compete with pioglitazone for selective and saturable binding to mitochondria.[63]

The next generation of TZDs mainly refers to MSDC-0160 and MSDC-0602, the potential drugs in treating neurodegenerative diseases,[65,66] and NASH,[67] respectively. MSDC-0602 was chosen from a medicinal chemistry program designed to significantly reduce the ability to bind and activate PPARγ while preserving the activity of a mitochondrial target (later found to be MPC).[59] In diet-induced obesity (DIO) and ob/ob animal models, Chen et al found that the pharmacological effects of MSDC-0602 were comparable to those of first-generation TZDs, pioglitazone, and rosiglitazone.[68,69] MSDC-0602 had an asymmetric carbon at the 5-position of the TZD ring like the first-generation TZDs mentioned. The carbonyl part of MSDC-0602 is a pre-chiral center (►**Fig. 8**). However, the changes in its carbon backbone reduce its capacity to bind PPAR. With free acid, MSDC-0602 was first developed.[69] The amount of exposure required to obtain a similar reduction in blood glucose as 45 mg of pioglitazone was assessed by a 3-month preclinical toxicology study and a Phase 2a trial involving participants with type 2 diabetes.[70] MSDC-0160, a TZD structurally similar to pioglitazone, is created similarly to MSDC-0602 and has comparable properties.[41] MSDC-0602K may inhibit HSC activity by promoting lipid oxidative catabolism, reducing glycolipid synthesis, to slow down the progression of NASH and reverse liver fibrosis. However, the MSDC-0602K-treated group did not meet the primary clinical endpoint, i.e., unsatisfactory improvement of NASH liver histology, in a phase IIb clinical trial, but in patients with T2DM combined with liver injury, MSDC-0602K demonstrated its safety and potential efficacy by detecting liver injury and glucose metabolism in a noninvasive manner. A phase III clinical trial of MSDC-0602K for glycemic control and

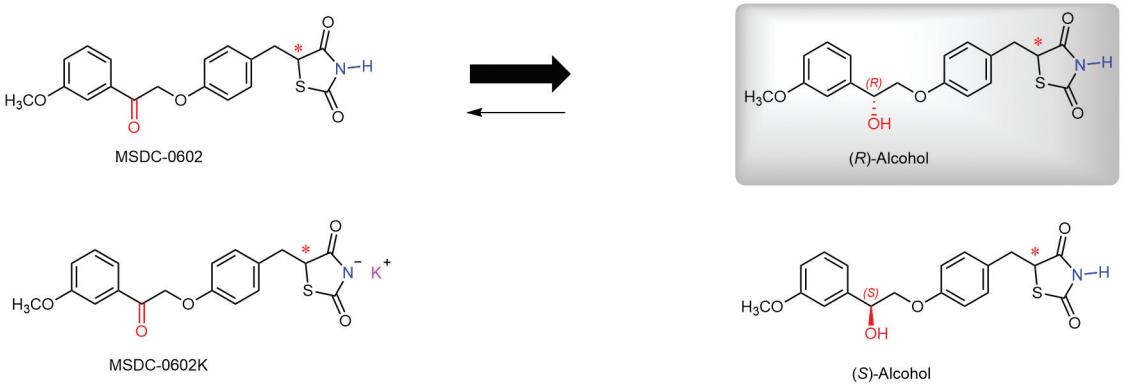


Fig. 8 Diastereomer of MSDC-0602 and MSDC-0602K including alcohols, accordingly.[69] In both humans and preclinical animals, MSDC-0602 is degraded to the (*R*)-alcohol, and even (*R*)-alcohol can be oxidized back to carbonyls, the equilibrium favors the alcohols. The asterisk denotes asymmetric carbon in all TZDs. TZDs, thiazolidinediones.

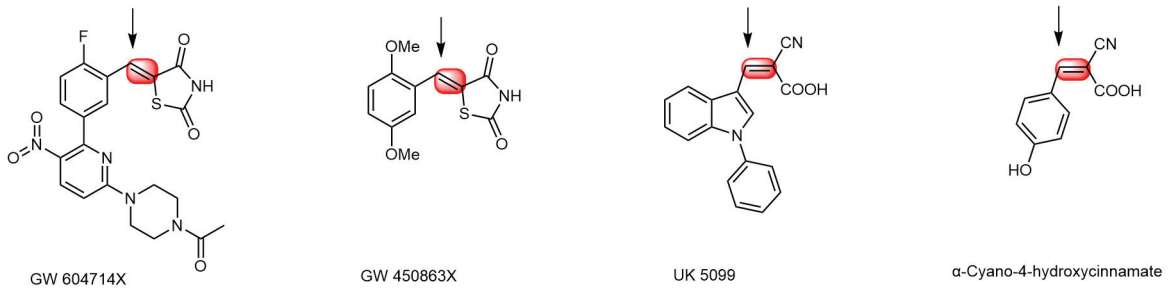


Fig. 9 Cyanate derivatives of GW604714X and GW450863X, and their chemical structures. The arrows show potential locations for the Michael addition reaction of sulfhydryl groups with activated double bonds. (Reprinted with permission from Hildyard JCW, Ämmälä C, Dukes ID, Thomson SA, Halestrap AP. Identification and characterization of a new class of highly specific and potent inhibitors of the mitochondrial pyruvate carrier. Biochim Biophys Acta BBA - Bioenerg 2005;1707(2-3):221–230.[43])

cardiovascular risk in patients with NASH combined with T2DM is currently underway.

TZDs lack selectivity because most TZDs bind and activate the peroxisome PPARγ.[54] Two novel thiazolidine compounds, GW604714X and GW450863X, in addition to being strong thiazolidine inhibitors with potencies similar to UK-5099, can influence the K^+/ATP channel function that plays a role in inhibiting potassium import.[43] The chemical structures of MPC inhibitors CHC and UK5099 are quite different from GW604714X and GW450863X. Notably, the thiazolidines lack a pyruvate-like carboxylic acid group. On the other hand, a double bond may be sufficiently reactive to undergo a Michael addition with a thiol group, as shown by the arrows in ►**Fig. 9**–[43] as the double bond in UK5099 and CHC.[42] The effect of mercaptoethanol on the absorbance spectra of two new inhibitors was determined by Hildyard et al, to test this possibility directly. The addition of 50 mmol/L H-mercaptoethanol resulted in the near vanish of the absorbance peak at approximately 350 nm. Thus, although the structures of the two inhibitors are very different, it is likely that they both act by targeting specific cysteine residues on the MPC.[43]

Considering the more hydrophobic potency of α-cyanate derivatives, GW604714X has a stronger affinity than GW450863X, which may be due to its greater hydrophobicity.[9] These new inhibitors may attach to distinct locations than cyanate salts because they do not appear to have any structural similarity to the monocarboxylates. They do, however, have an active double bond that can react with sulfhydryl groups, just as cyanate esters do. Another characteristic of the new inhibitors is that, like the α-cyanate derivatives, they take time to reach their maximum inhibition, and their potency and latency increase with increasing hydrophobicity. This may be due to the presence of hydrophobic binding pockets on the matrix surface of the carrier. In this respect, they are more selective than cyanate esters.[43]

Quinolones

7ACC1 and 7ACC2 are carboxycoumarin inhibitors of the monocarboxylate transporter 1 (MCT1), derivatives of coumarins, and are also known to inhibit MPC ($IC_{50} = 27 \pm 13$ μmol/L).[7] Studies has been shown that 7ACC2 is a potent MPC inhibitor, preventing both uptake and utilization of extracellular lactic acid by cancer cells, and also blocking the mitochondrial transport of pyruvate. 7ACC2 has been shown to interact directly with MPC and appears to be as effective as the prototype MPC inhibitor UK-5099.

Inhibition of MPC activity induces cytotoxicity, glycolysis stimulation, and noncompensatory inhibition of mitochondrial respiration. The reduction in hypoxia with 7ACC2 treatment suggests that tumor xenografts are sensitive to radiotherapy.[71]

Hodges et al used reporter genes sensitive to pyruvate to screen chemical libraries of drugs,[58] and to select new MPC inhibitors for further study, a pharmacophore model was developed using the structure of 7ACC2. As a result, carsalam and six quinolones were found and proved to be novel MPC inhibitors (►**Fig. 10**).

These substances have the same Michael receptor and therefore share a pharmacophore with 7ACC2. The molecular structure of carsalam, a nonsteroidal anti-inflammatory medication, also possesses a similar chemical structure.[58]

Quinolones are commonly used antibiotics. One of the main targets of quinolones in bacteria is the topoisomerase IV–DNA cleavage complex.[72] Fluoroquinolones have been reported to cause hypoglycemia in the past.[73] Quinolone-induced hypoglycemia is a side effect of these antibiotics, especially when used together with other antidiabetic drugs.[74] Interestingly, these MPC inhibitors with new chemical structures may ameliorate metabolic disorders in DIO mice, probably by inhibiting liver glucose production.[58] Despite being weak MPC inhibitors, recently developed drugs are easily chemically optimized and can be used to modify the structural makeup of compounds to increase their potency and selectivity toward MPC.

Others

A number of substances, such as the antibiotic nitrofurantoin, the cGMP-specific phosphodiesterase (PDE) inhibitor zaprinast,[75] and the antitumor medication lonidamine,[76] have also been demonstrated to inhibit MPC. Silibinin (an adjuvant for chronic hepatitis and cirrhosis) and valproic acid (a drug used to treat epilepsy, bipolar disorder, and migraine) have also been associated with inhibition of pyruvate uptake.[77]

Zaprinast was synthesized when studying xanthine as a possible antiallergic compound,[78] an inhibitor of cGMP-

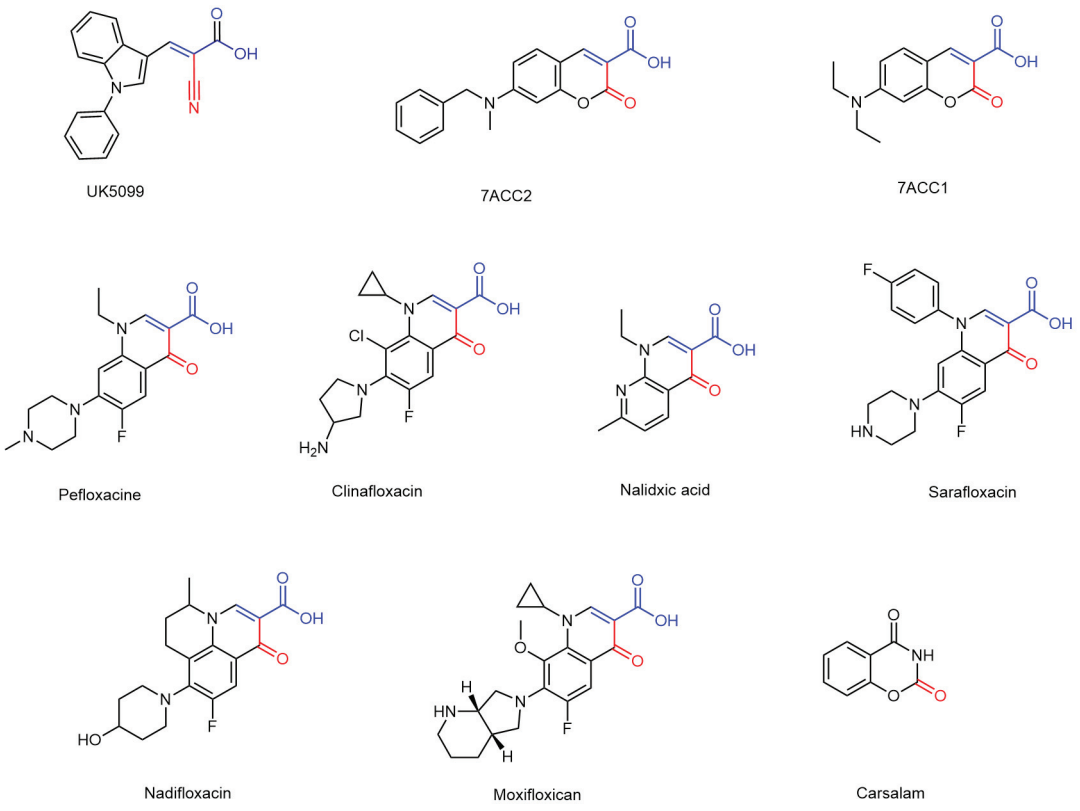


Fig. 10 Hodges et al screened the molecular structures of substances in the Pharmakon 1600 collection with comparable chemical structures and recognized MPC inhibitors.[58]

specific PDE, and a lead compound in the development of sildenafil (Viagra). Recent studies have shown that it inhibits pyruvate-driven oxygen consumption in brain mitochondria and pyruvate uptake in liver mitochondria.[49] Using mitochondria isolated from heart-specific *Mpc2*-deficient mice,[10] zaprinast has been shown to inhibit only pyruvate-mediated respiration in MPC-expressing mitochondria.[58] In addition, zaprinast was also found to be an effective MPC inhibitor in a study designed to use zaprinast to simulate the effect of PDE6 dysfunction on a model of retinal degeneration *in vitro*.[75] Although zaprinast can be used as a useful tool compound, its potential off-target effect, poor solubility, short half-life, and other drawbacks limit its research and make it difficult to promote the development of therapeutic drugs.[58]

Lonidamine was first introduced as an antitumor drug in 1979.[76] Reduced mitochondrial-activated protein activity is associated with the progression of several cancers due to its role in the Warburg effect. Recently, the inhibition of mitochondrial pyruvate metabolism has been identified as one of the mechanisms of action of long-acting lonidamine, an antitumor drug used to make tumors sensitive to chemotherapy and radiotherapy.[76] It inhibits pyruvate uptake in isolated mitochondria and suppresses the low-affinity MCT.[76,79] This confirmed the importance of mitochondrial pyruvate metabolism for cancer cell proliferation.[54]

Tavoulari et al compared the macromolecular interaction characteristics of pyruvate as a substrate with the MPC inhibitors UK5099, zaprinast, lonidamine, and mitoglitazone.[49] By using structural alignment to create a pharmacophore model and visualizing all shared pharmacophore properties, it was found that all inhibitors have three hydrogen-bonded acceptor groups that are tightly spaced (less than 2.95 Å) and are also present in the substrate pyruvate. This suggests that the binding effect of inhibitors and pyruvate in the MPC substrate binding bag is comparable. Additionally, all inhibitors have a central hydrophobic aromatic ring that is absent from pyruvate. Another common feature of certain inhibitors is the presence of an ionizable group that is negatively charged (such as lonidamine).[49]

Tavoulari et al created a pharmacophore model that incorporates all of the features that lead to an increase in compound **7** affinity to investigate whether this model could be used to identify off-target effects of existing small-molecule drugs on MPC. These features include an additional aromatic structure and two hydrogen-bonded receptors. Thus, two widely used substances that have been shown to interfere with mitochondrial function in the past have been identified as efficient MPC inhibitors (►**Fig. 11**).[80–82] The first is nitrofurantoin, an antibiotic used to treat bladder infections; the other is entacapone, used in combination therapy for PD. The lack of the middle aromatic ring part of nitrofurantoin may be the reason for its lower potency than entacapone. After the hydrogen bond receptor, the middle aromatic ring component appears to be the second requirement for MPC inhibition. It isolates inhibitors from substrates but is common to all inhibitors. Thus, the proposed basic chemical characteristics can direct the search for novel or unidentified MPC small-molecule inhibitors.[49]

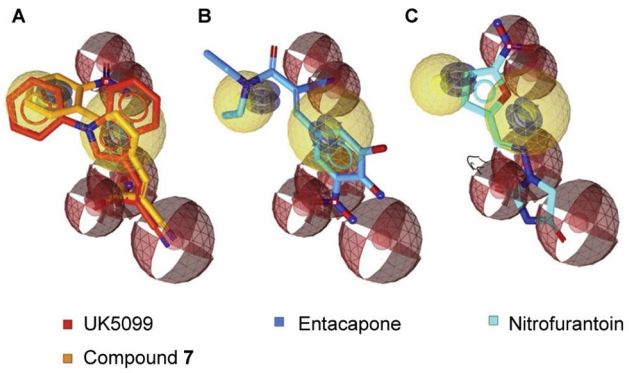


Fig. 11 (A) Pharmacophore of compound **7** and comparison with UK5099. The red ball represents the hydrogen receptor, the yellow ball represents the hydrophobic ring feature, the red star represents the negative ionization feature, and the blue ring represents the aromatic ring feature. (**B**) Fitting of entacapone in the expanded pharmacophore. (**C**) Fitting of nitrofurantoin to the extended pharmacophore. (Reprinted with permission from Tavoulari S, Schirris TJJ. Key features of inhibitor binding to the human mitochondrial pyruvate carrier hetero-dimer. Mol Metab 2022;60:101469.[49])

Conclusion and Perspectives

MPC, an important crossroad of intermediary metabolism, is inhibited in many diseases, such as diabetes mellitus, hair loss, tumors, CNS degenerative diseases, NASH, alopecia areata, etc. MPC has attracted wider attention as a potential drug target for metabolic diseases. Despite many studies on MPC inhibition, there are currently no marketed MPC inhibitors.

MPC inhibitors may develop off-targeting and drug resistance, due to their broad association with several disorders, particularly those of metabolism. Off-target may lead to toxicity, so more pharmacological and toxicological research is required to detect off-target tendencies early in the drug development process. In addition, additional clinical evidence is required to validate the specific diseases that MPC inhibitors can effectively treat. It was found that cells deficient in MPC1 undergo metabolic reprogramming to aerobic glycolysis, produce less ATP, migrate more, and become resistant to radiation and chemotherapy.[83] In a hypoxic environment, cancer cells exhibit strong glycolytic activity and are less sensitive to typical anticancer treatments. Inhibition of glycolysis efficiently kills colon cancer cells and lymphoma cells, and inhibition of glycolysis-induced ATP depletion also successfully triggered apoptosis in multidrug-resistant cells.[84] To date, most MPC inhibitors are compounds with known targets that have MPC inhibitory activity but lack specificity. No significant progress has been made in the study of its specificity, and in addition, the development of rational receptor-based drugs is challenging because the crystal structure of MPC proteins is still unknown. With the continuous development of small molecule and protein eutectic analysis technology, the analysis of MPC proteins may bring great improvement to the design of MPC inhibitors. In addition, computer-aided drug design helps in the discovery and optimization of lead compounds, aiding the discovery of novel MPC inhibitors from virtual screening, protein structure prediction, ADMET (absorption, distribution, metabolism, excretion, and toxicity) prediction, etc. For example, some proteins whose three-dimensional (3D) structures are unresolved can be assessed using protein structure prediction methods, including ab initio calculations and statistical methods. Meanwhile, in the case where the 3D structure of the target is unknown, a series of ligands with similar structure, the same type of action, and different sizes of activity can be analyzed, and the active site action of the matched target proteins can be investigated in reverse to find the common pharmacophore. Such drug design methods mainly include ligand-based pharmacophore modeling, virtual screening based on ligand similarity, and 3D-quantitative SAR. Many MPC inhibitors with different structural types have been discovered through different routes including computer-assisted drugs, which greatly enrich their structural types and provide new ideas for future compound design. In addition, the study of protein crystal structures through X-ray crystallography, nuclear magnetic resonance techniques, and electron microscopy can further contribute to the design and development of MPC inhibitors.

In conclusion, the study of the physiological function of MPC and its relationship with the mechanism of metabolic disorders may provide new ideas and therapeutic targets for the diseases. Similarly, summarizing the research progress of known small molecules of SAR and MPC inhibitors will also provide guidance and reference for the development of MPC inhibitors as possible target drugs in the future.

Funding

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Conflict of Interest

None declared.

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