Title ：Is “STAT3-CypD-PCr”a survival stable triangle in mitochondria?

Without Brief Title

Authors' names

Wu Liu(a, PhD), Eskandar Qaeda(a, PhD), Zeyao Tang(a, PhD,*);

(a)Acad integrated Med & collage of pharmacy, Department of pharmacy, Dalian Medical University, Dalian 116044, China.

*Correspondence: Zeyao Tang prof, Tel.: +86(411)86110412;

E-mail addresses: tangzeyao411@dmu.edu.com.

the Conflict of Interest

Ethical approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Availability of supporting data

Not applicable.

Competing interests

The authors declared no competing interests.

List of Abbreviations

ANT: Adenine-nucleotide translocator;
CypD: Cyclophilin D;
HF: Heart failure;
JAK / STAT: Janus protein tyrosine kinase / Signal transducer and transcription activator ;
MPT: Mitochondrial permeability transition;
PCr: Phosphocreatine;
PTP: Permeability transition pore;
ROS: Reactive oxygen species;

Text

As we know, mitochondria are called the metabolic hub of the cell and the key integrator of energy, maintaining the homeostasis of active oxygen, the homeostasis of Ca^{2+} and the internal management of mitochondrial dynamics. On the other side, a key feature of HF is characterized by the imbalance between ATP demand and production, occurred in mitochondria. Therefore, the over-generation of reactive oxygen species (ROS) from damaged mitochondria may cause oxidative stress, leading to the changes not only in ion homeostasis and but also in excitation- contraction coupling in failed cardiomyocytes.

1.The relationship between mPTP and CypD

During the mitoflash of oxidative stress, if the ROS level reaches a critical threshold, the membrane potential would be depolarized, and mPTP would act as a "safety valve" to stop the event[1]. The short-term transient mPTP opening is a physiological 'reset' adaptation mechanism of mitochondria, preventing mitochondrial Ca^{2+} overload. Moreover, a small amount of Ca^{2+} can not only stimulate mitochondrial metabolism, but also promote the production of ATP. However, when Ca^{2+} exceeds the threshold level and ATP is consumed in large amounts, voltage and Ca^{2+} sensitive channels will inhibit the respiratory complexes, subsequently trigger the opening of the permeability transition pore complexes, and eventually lead to mitochondrial permeability transition (MPT).

Mild MPT can be reversed by cyclophilin D (CypD) inhibitors, which acts as a positive regulator. Its action is to maintain a physiological metabolic homeostasis through the regulation of Ca^{2+} release via the modulation of the frequency of mitoflash and ATP synthase[2]; As we know, ATP synthase initiates its work in reverse to accelerate the pumping of protons out of the matrix at the expense of ATP, which antagonizes the reduction of membrane potential and promotes high matrix phosphate levels. However, uncontrolled ATP synthase is inefficient in replenishing membrane potential, which actually accelerates cell death. Besides, CypD can interacts with ATP synthase as a result of reducing the catalytic activity of ATP synthase by about 30%, subsequently reduces energy dissipation, and then dynamically relaxes the formation of ATP synthase c subunits through the mediation of conformational changes, eventually decreases the sensitivity and weakens the progress of mPTP[3].

It has been reported that CypD regulates energy homeostasis by enhancing the stability of Complex III, helping the assembly of supercomplexes to increase coupling. In addition, oxidative damage makes the protein fold abnormally. After CypD binds with chaperone, it blocks the conductance through these misfolded protein clusters. However, when the number of protein clusters exceeds the capacity that the chaperone is able to block their conductance, these misfolded protein clusters are not be "regulated" continuely. Afterwards, the openness of hole occurs, while mPTP is opened with high conductivity. Because of energy collapse, mitochondrial membrane potential collapse and bioenergy failure, swelling induced by the destruction of mitochondrial ion and solute balance, and the damage of cardiolipin cell membrane and the release of apoptotic factors, MPT is initiated in a complete high-conductivity, causing a critical Ca^{2+} concentration to a "death point". After that, the dual-acting CypD promotes the conformational change of adenine-nucleotide translocator (ANT) triggered by Ca^{2+}, leading to the opening of mPTP. Hence, we believe that CypD is the central transmission link of mitochondrial function, which is involved in regulating energy balance. Moreove, as a "gatekeeper" of amino acid, CypD also is a key regulator to maintain the plasticity of synthetic enzymes.

2. CypD restricted by STAT3

The Janus protein tyrosine kinase/signal transducer and activator of transcription (JAK/STAT) signaling pathway plays an important role in regulating cardiovascular disease homeostasis. STAT3 interacts with mitochondrial complex I and II to regulate the relevant proteins. The active kinases enhances the activity of the Electron

Transport Chain (ETC), increases the production of ATP and reduces the generation of ROS. The relationship between STAT3 and CypD is likely to be a dynamic interaction. Although it plays a vital role in the early stages, its importance may decrease over time. However, they complement each other and antagonize fine regulation[4]. Our previous study showed that PCr increased the expression of p-JAK through the regulation of JAK2/STAT3 signaling pathway, revealing the important role of activated p-JAK and p-STAT3, which have sufficient inhibitory action to keep mPTP close, and revive the function of ETC proteins, leading to the prevention of cardiomyopathy from apoptosis[5]. Our results, to some extent, prove and explain the phenomena mentioned in Rong Tian et al' article. i.e., "Cardiac magnetic resonance spectroscopy has shown that stores of highenergy creatine phosphate are decreased in association with oxidative modifications in ETC proteins[6].

3.We propose a hypothesis that PCr would be the cornerstone of survival

The disbalance between energy supply and demand is a pathological feature of HF, meaning that HF is a bioenergy hungry disease. Therefore, one of the ideas of new therapeutic strategies maybe is to develop a hemodynamically neutral drug that can target the myocardium. What we are more interested is to explore the non-energy mechanism of the treatment of this disease, sun as PCr. One kind of pharmacological action seems to provide energy, however, our previous large number of studies have confirmed that its role seems be more non energy mechanism, such as to produce antioxidant, anti-inflammatory through protecting the mitochondrial pathway[5].

In addition , it has been reported that PCr has a beneficial effect on all-cause short-term mortality including chronic HF[7]. Hence, We speculate that it may be related to the physicochemical properties of PCr or to the following mechanisms. PCr is smaller than ATP molecule and less polar, so it spreads more easily and has good compatibility, and it is easy to accumulate to higher levels in cells. PCr maintains the adenosine triphosphate pool and stabilizes the cell membrane conformation. Due to the electrostatic interaction with phospholipids, it reduces the production of lysophosphoglycerides in the hypoxic heart. The contact points between PCr and cell membrane also play a role in mechanically stabilizing the structure of the mitochondrial membrane. The prevention of cardiolipin oxidation and membrane lipid peroxidation is an essential to maintain the functional positioning of the ETC complex and supercomplex in the mitochondrial inner membrane[7], moreover, to maintain membrane fluidity, the integrity of ETC, the proton power of the electrical and chemical gradients and the proton circuit, resulting in allowing electrons to flow along the respiratory chain, which is the core of mitochondrial bioenergy and cell homeostasis.

Therefore, if PCr is provided sufficiently in external environment, CypD could play its core role better. On the other side, STAT3 limits or restricts the initiation of

CypD below the threshold so as not to induce the openness of mitochodrial pores completely.

Thereby, we propose a hypothesis, in summary, that the formation of an iron triangle among “STAT3-CypD-PCr” would maintain the survival of mitochondria (See Fig.1). We believe that PCr may be a new potential agent for the treatment of HF through the restoration of heart power in an unconventional anti-HF mechanism, providing new ideas for the regulation of cell survival, meanwhile revealing new therapeutic target for the treatment with HF and other relevant myocardial diseases.

Referece

1. Lan Wei-LaPierre, Alina Ainbinder, Kevin M.Tylock, Robert T. Dirksen. Substrate- dependent and cyclophilin D-independent regulation of mitochondrial flflashes in skeletal and cardiac muscle.Archives of Biochemistry and Biophysics. 2019, 665 (11) :122–131 .
2. David A Brown, Justin B Perry, Mitchell E Allen, Hani N Sabbah, Brian L Stauffer, Saame Raza Shaikh, John G F Cleland, Wilson S Colucci, Javed Butler 7 , Adriaan A Voors, Stefan D Anker, Bertram Pitt, Burkert Pieske, Gerasimos Filippatos, Stephen J Greene, Mihai Gheorghiade.Expert Consensus Document: Mitochondrial Function as a Therapeutic Target in Heart Failure. Nat Rev Cardiol. 2017;14(4):238-250.
3. Gisela Beutner, Ryan E. Alanzalon & George A. Porter Jr.Cyclophilin D regulates the dynamic assembly of mitochondrial ATP synthase into synthasomes. Sci Rep. 2017;7(1):14488.
4. Meier JA, Hyun M, Cantwell M, Raza A, Mertens C, Raje V, Sisler Tracy E, Torres-Odio S, Gispert S, Shaw PE, Baumann H, Bandyopadhyay D, Takabe K, LarnerAC. Stress-induced dynamic regulation of mitochondrial STAT3 and its association with cyclophilin D reduce mitochondrial ROS production. Sci Signal. 2017, 10(472):eaag2588.
5. Eskandar Qaed, Jiaqi Wang,Marwan Almoiliqy, Yanlin Song, Wu Liu, Peng Chu, Sawsan Alademi, Maria Alademi, Hailong Li, Mohammed Alshwmi,Mahmoud Al-Azab,Anil Ahsan, Samar Mahdi,Guozhu Han,Mengyue Niu,Amr Ali,Abdullah Shopit ,Hongyan Wang, Xiaodong Li, Abdullah Qaid, Xiaodong Ma, Tong Li,l Jinyong Peng ,Jing Ma,Jianbin Zhang, Zeyao Tang.Phosphocreatine Improves Cardiac Dysfunction by NormalizingMitochondrial Respiratory Function through JAK2/STAT3Signaling P athway In Vivo and In Vitro.Oxidative Medicine and Cellular Longevity. 2019, 30; 2019:6521218
6. Ivan Luptak, Aaron L Sverdlov, Marcello Panagia, Fuzhong Qin, David R Pimentel, Dominique Croteau, Deborah A Siwik, Joanne S Ingwall, Markus M Bachschmid, James A Balschi , Wilson S Colucci,. Decreased ATP production and myocardial contractile reserve in metabolic heart disease.J Mol Cell Cardiol. 2018;116:106-114.
7. Ben Zhou, Johannes Kreuzer, Caroline Kumsta, Lianfeng Wu, Kimberli J. Kamer,

Lucydalila Cedillo,Yuyao Zhang, Sainan Li,Michael C. Kacergis,Christopher M. Webster, Geza Fejes-Toth,Aniko Naray-Fejes-Toth, Sudeshna Das,Malene Hansen, Wilhelm Haas,and Alexander A. Soukas1, Mitochondrial Permeability Uncouples Elevated Autophagy and Lifespan Extension. Cell 2019,177(4):299-314.