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Review Article

Sepsis-induced Cardiac Mitochondrial Damage and Potential Therapeutic Interventions in the Elderly

Qun S. Zang*, Steven E. Wolf and Joseph P. Minei

Departments of Surgery, University of Texas Southwestern Medical Center, 5323 Harry Hines Blvd, Dallas, TX 75390, USA

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ABSTRACT: The incidence of sepsis and its attendant mortality risk are significantly increased with aging. Thus, severe sepsis in the elderly is likely to become an emerging concern in critical care units. Cardiac dysfunction is an important component of multi-organ failure after sepsis. In our laboratory, utilizing a pneumonia-related sepsis animal model, our research has been focused on the mechanisms underlying sepsis-induced cardiac failure. In this review, based on findings from others and ours, we discussed age-dependent decay in mitochondria and the role of mitochondrial reactive oxygen species (mtROS) in sepsis-induced cardiac inflammation and autophagy. Our recent discovery of a potential signal transduction pathway that triggers myocardial mitochondrial damage is also discussed. Because of the significance of mitochondria damage in the aging process and in sepsis pathogenesis, we hypothesize that specific enhancing mitochondrial antioxidant defense by mitochondria-targeted antioxidants (MTAs) may provide important therapeutic potential in treating elder sepsis patients. In this review, we summarized the categories of currently published MTA molecules and the results of preclinical evaluation of MTAs in sepsis and aging models.

Key words: mitochondria, sepsis, cardiac function, inflammation, autophagy, mitochondria-targeted antioxidants

Sepsis in Older Patients

Severe sepsis, defined by the presence of acute organ dysfunction, is the leading cause of death in intensive care units (ICUs) [1, 2] and the tenth leading cause of death overall in the US[3]. Despite improvements in antibiotic therapies and critical care techniques[4], approximately 215,000 Americans still die from sepsis each year [5].

In recent years, the number of older patients being admitted to ICUs has increased significantly [6]. Elderly patients, age over 65-year-old, account for about 60% of severe sepsis cases [7]. Increased incident in sepsis is 20% more in the elder population compared to younger counterparts [8], and the mortality rates of severe sepsis increase dramatically with aging [9]. In addition, among sepsis survivors, substantial and persistent cognitive impairment and functional disability are found to associate with aging, reported by a recent nationally prospective cohort study [10]. Such consequence will unavoidably result in a significant increase in the overall health burden of sepsis. The growth in the number of older sepsis patients can be explained by a decline in mortality rates that lead to increased life expectancy due to advances in modern medicine. It is expected that the population of elderly will grow more rapidly than any other age groups in the near future. Thus, the care for older patients present an emerging challenge for the clinical management of sepsis.

A number of aging-associated risk factors need to be considered when dealing with the treatment of elder sepsis patients. One major factor is age-related decline in immunity [11]. In elderly population, functions of T- and

***Correspondence should be addressed to:** Q. S. Zang, Ph. D, Department of Surgery, UT Southwestern Medical Center, 5323 Harry Hines Blvd., Dallas, TX 75390-9160, USA. Email: qun.zang@utsouthwestern.edu

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B- cells are defective [12, 13], notably by the loss of T- and B- cell repertoire [13, 14]. However, the elements of innate immunity, such as neutrophils, monocytes, macrophages, natural killer cells and dendritic cells, are well preserved [15-17]. Meanwhile, the cytokine expression is highly viable [18, 19], and the induction of pro-inflammatory cytokines takes prolonged period, reflecting progressive difficulties to meet the need of clearing microbial pathogens [20]. Because overwhelming inflammation is a characteristic response in sepsis, age-related immune deficiencies render the older patients at excess risk for the progression to severe sepsis after infection and acute injuries. Another physiological change in older patients that can not be ignored is the decline of nutrition status, which is caused by a number of reasons, including age-associated decrease of olfactory sensation, inactivity, social isolation, depression, poor dentition and chronic disease conditions [21]. In addition, pre-existing chronic comorbid medical conditions, such as HIV, cancer, diabetes and obesity, increase critical risk for older sepsis patients. Further, pre-admission functional status has been found to be an independent predictor for outcomes in older patients [22, 23]. Poor functional status is probably caused by disuse atrophy, lose of responsiveness to tropical hormones, neurological alterations and decrease in metabolism and dietary intake. Together, the deteriorated heath conditions are responsible for weakening an already compromised immune defense in the elderly.

Mitochondrial Damage and Aging Hearts

Mitochondrial dysfunction is a major focus in the study of aging process. The free radial theory of aging proposed by Harman half century ago suggests that aging is a result of deleterious effects of accumulation of harmful reactive oxygen species (ROS) [24]. Multiple intracellular sites produce ROS, such as xanthine oxidase in cytosol [25], NADPH oxidase at membrane [26] and lipid oxidation in peroxisomes [27]. However, the majority of oxidative stress burden comes from mitochondria, where ROS are generated as by products during oxidative phosphorylation and ATP production [28]. Scavenging of mitochondrial ROS (mtROS) is achieved via enzymatic and non-enzymatic antioxidants. Mitochondrial antioxidant enzymes consist of glutathione peroxidase (GPx), catalase (CAT) and manganese superoxide dismutase (MnSOD) [29-31]. An imbalance between mtROS production and scavenging leads to accumulation of mtROS, which disrupt the function of mitochondrial proteins, lipids, and DNA through structural modifications and therefore alter multiple aspects of mitochondrial function [32, 33]. As an extension to the free radical theory, it is proposed that mtROS are the main cause of functional deficiencies associated with aging [34].

However, to date, the correlation between ROS and aging still remains controversial, since published studies have provided evidence in both supporting and against the free radical theory of aging. For example, in yeast, deletion of three mitochondrial antioxidant genes, *SOD1* (Cu, Zn superoxide dismutase, CuZnSOD), *SOD2* (manganese superoxide dismutase, MnSOD) and *CCS1* (Copper chaperone), shortened the life span enormously, suggesting the importance of antioxidant defense in maintaining longevity [35]. Consistently, in *Caenorhabditis elegans*, giving the wild-type worms small synthetic SOD or catalase mimetics extended their life span by a mean of 44% [36]. It was further shown in mice that mitochondria-specific overexpression of human catalase (mCAT mice) attenuated age-associated mitochondrial dysfunction [37], reduced oxidative damage and significantly increased life span [38]. However, on the contrary, in a transgenic mouse study, overexpression of cytosolic CuZnSOD, catalase, or combinations of either CuZnSOD and catalase, or CuZnSOD and MnSOD failed to provide any longevity benefit [39]. Also, in the yeast study mentioned above, deletion of the other known mitochondrial antioxidant genes (*TTR1*, *CCD1*, *GLO4*, *TRR2*, *TRX3*, *GRX5*, *PRX1*) had little effect on life span [35]. In according with this, antioxidant supplements so far tested in human did not provide any beneficial effect over a well-balanced diet [40]. Thus, it appears that ROS are not the sole determinant factor but may provide significant impetus to the aging process. Accumulating evidence has indicated the importance of reducing mitochondrial ROS in life span and cardiac health benefits.

In the heart, mitochondria comprise about 30% of myocardial volume [41]. Thus, the heart is especially prone to mitochondrial oxidative stress. Studies suggest that age-dependent cardiac mitochondrial damage is caused by over production of mtROS [42, 43]. It was shown that, in mCAT mice, mitochondria-specific overexpression of human catalase provided resistance to heart failure induced by pressure overload [44], angiotensin II and Galphaq overexpression [45]. These transgenic mice exhibited improved cardiac performance and decreased age-associated cardiac pathology, such as ventricular fibrosis, and enlargement of myocardial fiber size [46]. Consistently, over expression of another mitochondrial antioxidant enzyme, manganese superoxide dismutase (MnSOD), protected cardiac morphology and normalized contractility of cardiomyocytes in a type 1 diabetes model [47]. In addition, gene knockout of MnSOD impaired left ventricular functions and promoted heart hypertrophy with accompanying fibrosis and necrosis [48]. Taken all

together, studies from these transgenic and gene knockout animal models have provided direct evidence to support the critical role of mtROS in cardiac dysfunction and aging. These investigations also suggest that targeted defense against mtROS may become an effective therapeutic strategy in dealing with age-associated cardiac malfunction.

Mitochondrial Damage in Septic Hearts

Cardiac dysfunction is a vital component of sepsis-associated multi-organ failure [49-51]. Severe sepsis patients with cardiac dysfunction have significantly higher mortality compared with patients without this condition (70 *vs.* 20%) [52, 53]. Clinically, the degree of mitochondrial dysfunction is tightly linked to sepsis outcomes [54, 55]. Since heart is a mitochondria-rich organ, the role of mitochondrial damage in sepsis-induced cardiac failure has been receiving a significant attention. Current studies suggest that multiple aspects of mitochondrial dysfunction, such as impaired metabolism, altered energy generation, and elevated production of mtROS, contribute to sepsis-induced heart failure [56-58].

Mitochondria and cardiac inflammation

Excessive inflammation is a characteristic response during sepsis and a major cause of organ failure, such as in the heart. Inflammation is triggered not only by pathogen-associated molecular patterns (PAMPs), presented by foreign pathogens, but also by danger-associated molecular patterns (DAMPs), formed by endogenous molecules released from damaged tissues [59-61]. Immune cells recognize PAMPs and DAMPs via four families of pattern recognition receptors (PRRs), including Toll-like receptors (TLRs), nucleotide-oligomerization domain (NOD)-like receptors (NLRs), cytoplasmic caspase-recruiting domain (CARD) helicases such as RIG-I/MDA5 [59, 62, 63], and C-type lectin receptors (specific expression on dendritic and myeloid cells[64, 65]). Downstream inflammatory responses are activated through signalosome pathway, in which NF-B is negatively regulated by the IκB kinase (IKK) signalosome pathway[66, 67], and/or through inflammasome pathway, in which inflammatory caspase 1 and 5 are controlled by the inflammasomes [68-70].

Studies in recent years revealed that a significant amount of DAMPs are generated from mitochondria. The list of mitochondria-derived DAMPs includes mtROS, mitochondrial DNA (mtDNA) fragments [71], N-formyl peptides [72-74], ATP [75, 76] and cytochrome C [61, 77]. These molecules are released from broken mitochondria into circulating system during cell death and organ injury, initiating inflammatory responses through multifactorial pathways. For example, circulating mtDNA fragments isolated from the plasma from trauma patients are capable to trigger peripheral inflammation in animal models [78, 79]. In macrophages, mtROS are essential components for the activation of inflammasome NLRP3 [80]. Mitochondrial matrix protein MAVS is part of the mitoxosome to activate NF-κB during antiviral responses [81]. These mitochondria-involved mechanisms are most likely all related to mtROS over production, since mtROS cause mitochondrial functional deficiency and structural rapture via direct oxidation [82, 83], and thus release mitochondrial molecules into cytoplasm or the circulating system.

A similar paradigm may be applicable to cardiac inflammation during sepsis. Certain PRRs, receptors to PAMPs and DAMPs, are identified in the heart tissue or cardiomyocytes [84-86]. In animal models, pharmacological inhibition of caspase 1 [87] or small interfering RNA (siRNA) blockage of NF-κB expression [88] prevented heart failure, attributing the activation of both signalosome and inflammasome pathways to sepsis-mediated cardiac dysfunction. Studies from others and ours suggest that mitochondrial signaling indeed plays a significant role in provoking inflammation in myocardium [89, 90].

Our laboratory previously developed a pneumonia-related sepsis model in rats [91]. In this model, rats were infected with *S. pneumoniae* and sepsis symptoms were confirmed by positive blood cultures, pulmonary inflammation, lactic acidosis, and a fall in mean arterial blood pressure 24 hours post-infection [92-95]. Using this model, we demonstrated that sepsis impaired cardiac mitochondria, causing compromised membrane integrity, increased oxidative stress and decreased antioxidant defense [96]. Further, this sepsis-triggered mitochondrial damage occurred prior to cardiac inflammatory responses such as cytokine productions and NF-κB activation [96]. Infiltration of neutrophil [89], accumulation of mtDNA fragments and ASC (apoptosis-associated speck-like protein containing a carboxy-terminal CARD), an inflammasome component (unpublished results), were also observed in septic myocardium. We further showed that specific suppression of mtROS protected cardiac mitochondria, attenuated inflammation and improved heart function in the same sepsis animal model [89]. We hypothesisze that sepsis-induced mtROS and inflammation in myocardium are linked through a positive feedback-signaling network. In this scenario, in response to septic challenge, mtROS participate in inciting inflammation that further triggers additional increases of mitochondrial damage and mtROS overproduction, leading to downstream exacerbation of inflammatory responses. In fact, myocardial mtROS increase and mitochondrial damage induced through

inflammatory mediators have been previously reported using sepsis [97] and non-sepsis models [98]. Thus, specific targeting mtROS, such as using mitochondria-targeted antioxidants, in early sepsis stage may have a therapeutic potential to control the progression of mitochondrial dysfunction and inflammation in later severe sepsis stage.

Mitochondria and cardiac autophagy

Increase in autophagy, a lysosome-dependent mechanism of removing damaged proteins and organelles [99], associates with failing hearts[100-104]. Autophagy cascade is initiated by Beclin-1 (autophagy-regulated gene 6) [105], which forms complex with class III phosphoinositide 3-kinase (PI3K) to promote the formation of autophagosomes. After subsequent fusion with lysosomes, the materials inside autophagosomes are degraded [106, 107]. Autophagy is either protective or detrimental to myocardium, depending on varying disease conditions [101, 104, 108, 109]. It has been proposed that, under physiological responses or mild stress, autophagy provides cellular quality control to promote survival and is therefore adaptive. However, under severe or chronic stress, excessive or inadequate autophagy causes massive self-degradation or accumulation of toxic materials; both are maladaptive and eventually provoke cell death [110, 111].

As mentioned earlier, mitochondrial oxidative stress burden increases along with aging. Effective removal of damaged mitochondria and unwanted mitochondrial molecules is essential for maintaining a healthy heart. However, recent studies strongly indicate an age-associated impairment of cardiac autophagy [112, 113]. One possible factor for this autophagy deficiency lies in the enlargement of mitochondria. It was shown that mitochondria in aged cells are often enlarged, showing structural changes such as swelling, loss of cristae, and/or almost complete damage of mitochondrial components [114, 115]. A recent *in vitro* study in cultured neonatal cardiomyocytes suggests that autophagic turnover of small mitochondria is more efficient than that of the large ones[116]. Further, disruption of lysosomal function is another factor contributing to the slow-down autophagy in the aged hearts [117-119]. As a result of autophagy deficiency, aged hearts unavoidably accumulate damaged mitochondria, mtROS and other mitochondria-derived DAMPs molecules, which increase the heart vulnerability to deteriorative inflammatory and autophagic responses under trauma and sepsis conditions.

In sepsis, increase in autophagy has been detected in multiple organs, including the heart, in animal models and in clinical samples [56, 120-123]. However, the mechanism(s) underling its occurrence remains unclear. Function of mtROS in induction of autophagy has been suggested by studies from other disease conditions. For example, in a hypertensive cardiomyopathy model, angiotensin II-provoked autophagy was inhibited by overexpression of mitochondria-targeted antioxidant enzyme catalase [45]. In Hela cells, starvation-induced autophagy was decreased when mtROS failed to increase [124]. On the other hand, evidence also indicates that autophagy exerts a control over mtROS levels, since autophagy is often initiated in order to remove toxic molecules, including mtROS, under certain stress conditions [125, 126]. In our current preliminary investigation, we obtained data suggesting that mtROS may have a stimulatory role in sepsis-induced autophagic responses in the heart, and we hypothesize that, in septic hearts, imbalanced overproduction of mtROS starts an autophagy-promoting feed-forward pathway that leads to pathological progressions.

Current knowledge with regards to the role of cardiac autophagy in sepsis outcomes, adaptive or maladaptive, is still limited and inconclusive. In septic hearts, pharmacological activation of autophagy in mouse CLP model [127] or in cultured cardiomyocytes [128] suggests that stimulating autophagy is protective to myocardium, and thus autophagy is an adaptive response. However, a recent publication showed that reducing autophagy by an autophagy inhibitor or antioxidants improves cardiac contractility in a mouse lipopolysaccharide (LPS)-induced sepsis model [129], suggesting cardiac autophagy as a maladaptive response. The discrepancy of these observations is probably caused by the differences in the level of autophagy, the severity of sepsis and the timing of drug administration in individual experimental settings. Future investigations using autophagy transgenic and knockout models are needed to address the role of autophagy in septic hearts. So far, autophagy status in older sepsis patients remains unclear. Detailed analysis of cardiac autophagy in aging animal models in response to sepsis challenge will help us to understand the pathological conditions in aging septic hearts, assisting further improvement on therapeutic strategies to combat cardiac failure in elder sepsis patients.

Signal transduction of cardiac mitochondrial damage in sepsis

To date, little is known about the intracellular signal transduction pathway(s) that triggers mitochondrial damage in the heart after sepsis. Recent investigation from our laboratory suggests that sepsis alters mitochondrial translocation of tyrosine kinase cSrc and phosphatase SHP_2, which may stimulate mitochondrial dysfunction and mtROS production in myocardium[130].

During the past several years, a growing body of evidence has suggested that certain well-known intracellular signaling molecules, such Src-family tyrosine kinases [131], tyrosine phosphatases PTP-1B and SHP_2 [132], and serine/threonine kinases, protein kinase C (PKC) [133, 134] and extracellular-signal-regulated kinases (ERK) [135, 136], also provide important functions inside mitochondria. Their intra-mitochondria localization was verified using immune electron microscopy [131, 135, 137] and western blot analysis [131, 132]. However, since these molecules do not possess mitochondria-sorting peptide, the mechanism of their mitochondrial translocation has not been understood yet. In mitochondria, these kinases and phosphatases may play an important part in control of mitochondrial function and structure through reversible phosphorylation and dephosphorylation [132, 136, 138, 139]. Proteomic analysis of healthy mitochondria from rat brains [140] and from mouse hearts [141] captured phosphorylation sites on critical enzymes of mitochondria metabolism, membrane components and biosynthesis molecules. Some key components of oxidative phosphorylation (OXPHOS) complexes, such as subunits of NADH-coenzyme Q oxidoreductase (complex I) [142, 143], subunit IV of cytochrome c oxidase (complex IV) [144] and subunit ∂ of F0F1-ATP synthase (complex V) [145], have been identified as targets of phosphorylation. In addition to this category, other mitochondrial functional proteins, such as adenine nucleotide translocator 1 (ANT1)[138], aconitase [146] and telomerase reverse transcriptase (TERT)[147], were also shown regulated via tyrosine phosphorylation and dephosphorylation. It is noteworthy to point out that, since mtROS are generated from the reactions of OXPHOS complexes [148], changes in mitochondrial-localized kinases and phosphotases will inevitably affect the production levels of mtROS.

In a pneumonia-related sepsis animal model, we found that a significant decrease in mitochondrial Src and an increase in mitochondrial SHP_2 in myocardium were directly associated with sepsis [130]. Correlated with these changes, tyrosine phosphorylation of mitochondrial proteins, including some essential structural and functional proteins, was dramatically reduced. Both *in vitro* biochemical analysis and *in vivo* animal study suggest that OXPHOS complex I and III contain putative substrates of Src and SHP_2 [130], consistent with previous findings using small molecule inhibitors that implicated Src family kinases and SHP_2 phosphatase as main regulators of tyrosine phosphorylation in mitochondria [132, 149, 150]. We hypothesize that, during sepsis, certain receptors of pathogen-associated molecular patterns (PAMPs) and/or danger-associated molecular patterns (DAMPs) alter mitochondrial translocation of Src and SHP_2 in myocardium. The resulted changes in tyrosine phosphorylation of mitochondrial proteins produce functional deficiency and mtROS overproduction, and thus damaged mitochondria further generate more DAMPs to aggravate inflammatory responses and organ dysfunction [71]. However, several aspects need to be further addressed to support this hypothesis. Mitochondrial substrates of Src and SHP_2 remain to be defined, and the upstream receptor(s) that regulates mitochondrial translocation of Src and SHP_2 awaits to be identified. Furthermore, whether alteration of mitochondrial Src and SHP_2 relates to the production of mitochondrial-derived DAMPs to stimulate inflammation and how this signaling pathway affects cardiomyocyte function deserve further elucidation.

Current studies started to reveal some evidence that correlates the changes in reversible protein phosphorylation/dephosphorylation inside mitochondria with aging. For example, intra-mitochondrial AMP-activated protein kinase (AMPK) activity decreased with age, contributing to reduced mitochondrial biogenesis [151, 152]. Mitochondrial translocation of p66-Shc, an adaptor protein to tyrosine kinase receptors, stimulates mtROS production and regulates longevity in mice [153, 154]. Given the important role of kinases and phosphatase in the regulation of mitochondrial function, future research to understand the kinases and phosphatases events inside mitochondria will promote the understanding of pathogenesis in the heart of older sepsis patients. Research in this area will also help to identify new therapeutic targets to control cardiac dysfunction.

Therapeutic Approaches Targeting mtROS

For a long time, oxidative stress has been well recognized as a major promoter in sepsis pathogenesis [155, 156], and antioxidants are expected to attenuate inflammation and improve survival following sepsis. However, although this expectation has been met in animal sepsis models [157, 158], clinical trials of antioxidant therapies have led to inconsistent results [159-161]. One limitation of the conventional antioxidants is that they are globally acting agents, and insufficient dosage and/or lower efficacy are very likely to be the reasons for the failures [162].

Because ROS are mainly generated via mitochondrial respiration, mitochondria themselves are thought to be the primary target of oxidative damage. Targeting antioxidant defense specifically in mitochondria has been expected to provide more effective mitochondrial protection. Accordingly, strategies for mitochondria-targeted delivery of antioxidants are being developed [163-170]. One such approach covalently links bio-molecules to lipophilic triphenylphosphonium cation (TPP^+). Due to a positive charge, the molecules are driven by the mitochondrial membrane potential to accumulate solely in

mitochondria [167-169]. Another group of targeted antioxidants, Szeto-Schiller (SS)-peptides, are small positively charged peptides that accumulate in mitochondria independent of membrane potential[164, 170]. These novel mitochondria-targeted antioxidants (MTAs) have demonstrated their higher capability in various experimental settings to fight oxidative stress and to protect mitochondrial function [171-174].

Currently, MTAs have not yet been applied clinically. A clinical trial of mitochondrial-targeted ubiquinone (MitoQ) showed its benefit in treating liver inflammation [175], and a phase IIb human trial has been initiated in the U.K. to assess the efficacy of MitoQ in non-alcoholic fatty liver disease[176]. To date, the therapeutic potential of MTAs is under intense investigation using pre-clinical models of mitochondrial abnormalities-associated diseases such as neurodegenerative diseases [177, 178], cardiac dysfunction[179], cardiac ischemia-reperfusion injury [163], hypertension [180], diabetes [181], and sepsis [182, 183].

In sepsis animal models, MitoQ showed its therapeutic benefits in the improvement of cardiac function and prevention of liver damage [182, 183]. In a recent published study, we compared the effects of Mito-Vit-E with untargeted vitamin E in the rat pneumonia-related sepsis model [89]. Both types of antioxidants exhibited significant inhibition on peripheral and cardiac inflammation. At the same dose, Mito-Vit-E provided higher efficacy to reduce cytokine production and to impede neutrophil infiltration in myocardium. This advantage of Mito-Vit-E over vitamin E is likely caused by the fact that vitamin E is distributed globally and its protection of mitochondria against oxidative damage is less efficient, especially in mitochondria-enriched organs such as the heart. Further study of MTAs effects using different sepsis models will allow us to recommend possible candidate molecules for clinical studies and promote translating the application of these novel antioxidants into significantly improved clinical outcomes.

Studies of antioxidant SkQ1, TPP^+-conjugated plastoquinone [184], in animal models have revealed certain evidence to support using MTAs as an anti-aging approach. This compound reversed aging-dependent behavioral trait in rats after a ten-week-treatment[185]. In mice with lifelong treatment, SkQ1 significantly reduced age-related changes of hematopoietic and mesenchymal progenitor cells [186]. SkQ1 also showed effect to prolong lifespan in Drosophila [187], mice and hamsters [188]. It has been suggested that supplementation with low doses MTAs is a promising intervention to achieve a healthy aging. However, evidence from both pre-clinical and clinical research is needed to support this hypothesis.

Conclusion

Sepsis represents as a major threat in critical care units. Treatment for this deadly condition remains to be supportive care such as using intravenous fluids and oxygen [4]. Most current attempts of molecular target-based treatments have failed clinically [162, 189]. Even though older patients account nearly two third of severe sepsis cases [7], elderly population is likely to be excluded when new anti-sepsis and anti-microbial agents are tested in clinical trials. It is now realized that aging-associated decay in mitochondrial function and overproduction of mitochondrial oxidative stress are key elements to cause deficiencies in inflammation and autophagy, which are critical responses to trigger organ failure in severe sepsis stage. We anticipate that protection of mitochondria by mitochondria-targeted antioxidants (MTAs) may provide an effective therapeutic strategy for sepsis patients, especially for the elderly. Thus, future preclinical and clinical assessment of MTAs will have important translational implications to significantly impact patient care quality and clinical outcomes.

References

[1] Angus DC, Pereira CA, Silva E (2006). Epidemiology of severe sepsis around the world. Endocr Metab Immune Disord Drug Targets, 6: 207-212

[2] Bone RC, Balk RA, Cerra FB, Dellinger RP, Fein AM, Knaus WA, et al. (1992). Definitions for sepsis and organ failure and guidelines for the use of innovative therapies in sepsis. The ACCP/SCCM Consensus Conference Committee. American College of Chest Physicians/Society of Critical Care Medicine. Chest, 101: 1644-1655

[3] Moss M, Martin GS (2004). A global perspective on the epidemiology of sepsis. Intensive care medicine, 30: 527-529

[4] Levy MM, Dellinger RP, Townsend SR, Linde-Zwirble WT, Marshall JC, Bion J, et al. The Surviving Sepsis Campaign: results of an international guideline-based performance improvement program targeting severe sepsis. Crit Care Med, 38: 367-374

[5] O'Brien JM, Jr., Ali NA, Aberegg SK, Abraham E (2007). Sepsis. Am J Med, 120: 1012-1022

[6] Marik PE (2006). Management of the critically ill geriatric patient. Critical care medicine, 34: S176-182

[7] Angus DC, Linde-Zwirble WT, Lidicker J, Clermont G, Carcillo J, Pinsky MR (2001). Epidemiology of severe sepsis in the United States: analysis of incidence, outcome, and associated costs of care. Critical care medicine, 29: 1303-1310

[8] Martin GS, Mannino DM, Moss M (2006). The effect of age on the development and outcome of adult sepsis. Critical care medicine, 34: 15-21

[9] Nasa P, Juneja D, Singh O, Dang R, Arora V (2012). Severe sepsis and its impact on outcome in elderly and

very elderly patients admitted in intensive care unit. Journal of intensive care medicine, 27: 179-183
[10] Iwashyna TJ, Ely EW, Smith DM, Langa KM (2010). Long-term cognitive impairment and functional disability among survivors of severe sepsis. JAMA : the journal of the American Medical Association, 304: 1787-1794
[11] Franceschi C, Bonafe M, Valensin S (2000). Human immunosenescence: the prevailing of innate immunity, the failing of clonotypic immunity, and the filling of immunological space. Vaccine, 18: 1717-1720
[12] Miller RA (2000). Effect of aging on T lymphocyte activation. Vaccine, 18: 1654-1660
[13] Weksler ME (2000). Changes in the B-cell repertoire with age. Vaccine, 18: 1624-1628
[14] Grubeck-Loebenstein B, Wick G (2002). The aging of the immune system. Advances in immunology, 80: 243-284
[15] Plackett TP, Boehmer ED, Faunce DE, Kovacs EJ (2004). Aging and innate immune cells. Journal of leukocyte biology, 76: 291-299
[16] Sanchez M, Lindroth K, Sverremark E, Gonzalez Fernandez A, Fernandez C (2001). The response in old mice: positive and negative immune memory after priming in early age. International immunology, 13: 1213-1221
[17] Lebecque S (2000). Antigen receptors and dendritic cells. Vaccine, 18: 1603-1605
[18] Herrero C, Marques L, Lloberas J, Celada A (2001). IFN-gamma-dependent transcription of MHC class II IA is impaired in macrophages from aged mice. The Journal of clinical investigation, 107: 485-493
[19] Gabriel P, Cakman I, Rink L (2002). Overproduction of monokines by leukocytes after stimulation with lipopolysaccharide in the elderly. Experimental gerontology, 37: 235-247
[20] Bruunsgaard H, Skinhoj P, Qvist J, Pedersen BK (1999). Elderly humans show prolonged in vivo inflammatory activity during pneumococcal infections. The Journal of infectious diseases, 180: 551-554
[21] Jensen GL, McGee M, Binkley J (2001). Nutrition in the elderly. Gastroenterology clinics of North America, 30: 313-334
[22] Chernow B (1999). Variables affecting outcome in critically ill patients. Chest, 115: 71S-76S
[23] Stump TE, Callahan CM, Hendrie HC (2001). Cognitive impairment and mortality in older primary care patients. Journal of the American Geriatrics Society, 49: 934-940
[24] Harman D (1956). Aging: a theory based on free radical and radiation chemistry. J Gerontol, 11: 298-300
[25] Harrison R (2002). Structure and function of xanthine oxidoreductase: where are we now? Free radical biology & medicine, 33: 774-797
[26] Bedard K, Krause KH (2007). The NOX family of ROS-generating NADPH oxidases: physiology and pathophysiology. Physiological reviews, 87: 245-313
[27] Schrader M, Fahimi HD (2006). Peroxisomes and oxidative stress. Biochimica et biophysica acta, 1763: 1755-1766
[28] Barja G (1998). Mitochondrial free radical production and aging in mammals and birds. Annals of the New York Academy of Sciences, 854: 224-238
[29] Antunes F, Han D, Cadenas E (2002). Relative contributions of heart mitochondria glutathione peroxidase and catalase to H(2)O(2) detoxification in in vivo conditions. Free radical biology & medicine, 33: 1260-1267
[30] Melov S, Coskun P, Patel M, Tuinstra R, Cottrell B, Jun AS, et al. (1999). Mitochondrial disease in superoxide dismutase 2 mutant mice. Proceedings of the National Academy of Sciences of the United States of America, 96: 846-851
[31] Radi R, Turrens JF, Chang LY, Bush KM, Crapo JD, Freeman BA (1991). Detection of catalase in rat heart mitochondria. The Journal of biological chemistry, 266: 22028-22034
[32] Beal MF (2002). Oxidatively modified proteins in aging and disease. Free radical biology & medicine, 32: 797-803
[33] Croteau DL, Bohr VA (1997). Repair of oxidative damage to nuclear and mitochondrial DNA in mammalian cells. The Journal of biological chemistry, 272: 25409-25412
[34] Harman D (1972). Free radical theory of aging: dietary implications. The American journal of clinical nutrition, 25: 839-843
[35] Unlu ES, Koc A (2007). Effects of deleting mitochondrial antioxidant genes on life span. Annals of the New York Academy of Sciences, 1100: 505-509
[36] Melov S, Ravenscroft J, Malik S, Gill MS, Walker DW, Clayton PE, et al. (2000). Extension of life-span with superoxide dismutase/catalase mimetics. Science, 289: 1567-1569
[37] Lee HY, Choi CS, Birkenfeld AL, Alves TC, Jornayvaz FR, Jurczak MJ, et al. (2010). Targeted expression of catalase to mitochondria prevents age-associated reductions in mitochondrial function and insulin resistance. Cell metabolism, 12: 668-674
[38] Schriner SE, Linford NJ, Martin GM, Treuting P, Ogburn CE, Emond M, et al. (2005). Extension of murine life span by overexpression of catalase targeted to mitochondria. Science, 308: 1909-1911
[39] Perez VI, Van Remmen H, Bokov A, Epstein CJ, Vijg J, Richardson A (2009). The overexpression of major antioxidant enzymes does not extend the lifespan of mice. Aging cell, 8: 73-75
[40] Dolara P, Bigagli E, Collins A (2012). Antioxidant vitamins and mineral supplementation, life span expansion and cancer incidence: a critical commentary. European journal of nutrition, 51: 769-781
[41] Kayar SR, Banchero N (1987). Volume density and distribution of mitochondria in myocardial growth and hypertrophy. Respir Physiol, 70: 275-286
[42] Judge S, Jang YM, Smith A, Hagen T, Leeuwenburgh C (2005). Age-associated increases in oxidative stress and antioxidant enzyme activities in cardiac interfibrillar mitochondria: implications for the mitochondrial theory of aging. FASEB journal : official publication of the

Federation of American Societies for Experimental Biology, 19: 419-421
[43] Dai DF, Rabinovitch PS, Ungvari Z (2012). Mitochondria and cardiovascular aging. Circulation research, 110: 1109-1124
[44] Dai DF, Hsieh EJ, Liu Y, Chen T, Beyer RP, Chin MT, et al. (2012). Mitochondrial proteome remodelling in pressure overload-induced heart failure: the role of mitochondrial oxidative stress. Cardiovascular research, 93: 79-88
[45] Dai DF, Johnson SC, Villarin JJ, Chin MT, Nieves-Cintron M, Chen T, et al. (2011). Mitochondrial oxidative stress mediates angiotensin II-induced cardiac hypertrophy and Galphaq overexpression-induced heart failure. Circulation research, 108: 837-846
[46] Dai DF, Santana LF, Vermulst M, Tomazela DM, Emond MJ, MacCoss MJ, et al. (2009). Overexpression of catalase targeted to mitochondria attenuates murine cardiac aging. Circulation, 119: 2789-2797
[47] Shen X, Zheng S, Metreveli NS, Epstein PN (2006). Protection of cardiac mitochondria by overexpression of MnSOD reduces diabetic cardiomyopathy. Diabetes, 55: 798-805
[48] Loch T, Vakhrusheva O, Piotrowska I, Ziolkowski W, Ebelt H, Braun T, et al. (2009). Different extent of cardiac malfunction and resistance to oxidative stress in heterozygous and homozygous manganese-dependent superoxide dismutase-mutant mice. Cardiovascular research, 82: 448-457
[49] Court O, Kumar A, Parrillo JE (2002). Clinical review: Myocardial depression in sepsis and septic shock. Crit Care, 6: 500-508
[50] Zanotti-Cavazzoni SL, Hollenberg SM (2009). Cardiac dysfunction in severe sepsis and septic shock. Curr Opin Crit Care, 15: 392-397
[51] Rudiger A, Singer M (2007). Mechanisms of sepsis-induced cardiac dysfunction. Critical care medicine, 35: 1599-1608
[52] Blanco J, Muriel-Bombin A, Sagredo V, Taboada F, Gandia F, Tamayo L, et al. (2008). Incidence, organ dysfunction and mortality in severe sepsis: a Spanish multicentre study. Crit Care, 12: R158
[53] Parrillo JE, Parker MM, Natanson C, Suffredini AF, Danner RL, Cunnion RE, et al. (1990). Septic shock in humans. Advances in the understanding of pathogenesis, cardiovascular dysfunction, and therapy. Annals of internal medicine, 113: 227-242
[54] Brealey D, Brand M, Hargreaves I, Heales S, Land J, Smolenski R, et al. (2002). Association between mitochondrial dysfunction and severity and outcome of septic shock. Lancet, 360: 219-223
[55] Cairns CB, Moore FA, Haenel JB, Gallea BL, Ortner JP, Rose SJ, et al. (1997). Evidence for early supply independent mitochondrial dysfunction in patients developing multiple organ failure after trauma. The Journal of trauma, 42: 532-536
[56] Watts JA, Kline JA, Thornton LR, Grattan RM, Brar SS (2004). Metabolic dysfunction and depletion of mitochondria in hearts of septic rats. Journal of molecular and cellular cardiology, 36: 141-150
[57] Levy RJ (2007). Mitochondrial dysfunction, bioenergetic impairment, and metabolic down-regulation in sepsis. Shock, 28: 24-28
[58] Callahan LA, Supinski GS (2007). Diaphragm and cardiac mitochondrial creatine kinases are impaired in sepsis. J Appl Physiol, 102: 44-53
[59] Akira S, Uematsu S, Takeuchi O (2006). Pathogen recognition and innate immunity. Cell, 124: 783-801
[60] Matzinger P (1994). Tolerance, danger, and the extended family. Annu Rev Immunol, 12: 991-1045
[61] Codina R, Vanasse A, Kelekar A, Vezys V, Jemmerson R (2010). Cytochrome c-induced lymphocyte death from the outside in: inhibition by serum leucine-rich alpha-2-glycoprotein-1. Apoptosis, 15: 139-152
[62] Creagh EM, O'Neill LA (2006). TLRs, NLRs and RLRs: a trinity of pathogen sensors that co-operate in innate immunity. Trends Immunol, 27: 352-357
[63] Uematsu S, Akira S (2006). Toll-like receptors and innate immunity. J Mol Med, 84: 712-725
[64] Geijtenbeek TB, Gringhuis SI (2009). Signalling through C-type lectin receptors: shaping immune responses. Nature reviews. Immunology, 9: 465-479
[65] Osorio F, Reis e Sousa C (2011). Myeloid C-type lectin receptors in pathogen recognition and host defense. Immunity, 34: 651-664
[66] Orange JS, Geha RS (2003). Finding NEMO: genetic disorders of NF-[kappa]B activation. J Clin Invest, 112: 983-985
[67] Israel A (2000). The IKK complex: an integrator of all signals that activate NF-kappaB? Trends Cell Biol, 10: 129-133
[68] Cinel I, Opal SM (2009). Molecular biology of inflammation and sepsis: a primer. Critical care medicine, 37: 291-304
[69] Martinon F, Mayor A, Tschopp J (2009). The inflammasomes: guardians of the body. Annu Rev Immunol, 27: 229-265
[70] Lee MS, Kim YJ (2007). Signaling pathways downstream of pattern-recognition receptors and their cross talk. Annu Rev Biochem, 76: 447-480
[71] Zhang Q, Raoof M, Chen Y, Sumi Y, Sursal T, Junger W, et al. (2010). Circulating mitochondrial DAMPs cause inflammatory responses to injury. Nature, 464: 104-107
[72] Carp H (1982). Mitochondrial N-formylmethionyl proteins as chemoattractants for neutrophils. J Exp Med, 155: 264-275
[73] Czapiga M, Gao JL, Kirk A, Lekstrom-Himes J (2005). Human platelets exhibit chemotaxis using functional N-formyl peptide receptors. Exp Hematol, 33: 73-84
[74] Bjorkman L, Karlsson J, Karlsson A, Rabiet MJ, Boulay F, Fu H, et al. (2008). Serum amyloid A mediates human neutrophil production of reactive oxygen species through a receptor independent of formyl peptide receptor like-1. Journal of leukocyte biology, 83: 245-253
[75] Ghiringhelli F, Apetoh L, Tesniere A, Aymeric L, Ma Y, Ortiz C, et al. (2009). Activation of the NLRP3 inflammasome in dendritic cells induces IL-1beta-dependent adaptive immunity against tumors. Nature medicine, 15: 1170-1178

[76] Iyer SS, Pulskens WP, Sadler JJ, Butter LM, Teske GJ, Ulland TK, et al. (2009). Necrotic cells trigger a sterile inflammatory response through the Nlrp3 inflammasome. Proceedings of the National Academy of Sciences of the United States of America, 106: 20388-20393

[77] Pullerits R, Bokarewa M, Jonsson IM, Verdrengh M, Tarkowski A (2005). Extracellular cytochrome c, a mitochondrial apoptosis-related protein, induces arthritis. Rheumatology (Oxford), 44: 32-39

[78] Zhang Q, Itagaki K, Hauser CJ MITOCHONDRIAL DNA IS RELEASED BY SHOCK AND ACTIVATES NEUTROPHILS VIA p38 MAP-KINASE. Shock,

[79] Zhang Q, Raoof M, Chen Y, Sumi Y, Sursal T, Junger W, et al. Circulating mitochondrial DAMPs cause inflammatory responses to injury. Nature, 464: 104-107

[80] Zhou R, Yazdi AS, Menu P, Tschopp J (2011). A role for mitochondria in NLRP3 inflammasome activation. Nature, 469: 221-225

[81] Seth RB, Sun L, Ea CK, Chen ZJ (2005). Identification and characterization of MAVS, a mitochondrial antiviral signaling protein that activates NF-kappaB and IRF 3. Cell, 122: 669-682

[82] Tsutsui H, Kinugawa S, Matsushima S (2008). Oxidative stress and mitochondrial DNA damage in heart failure. Circ J, 72 Suppl A: A31-37

[83] Ballinger SW (2005). Mitochondrial dysfunction in cardiovascular disease. Free radical biology & medicine, 38: 1278-1295

[84] Boyd JH, Mathur S, Wang Y, Bateman RM, Walley KR (2006). Toll-like receptor stimulation in cardiomyoctes decreases contractility and initiates an NF-kappaB dependent inflammatory response. Cardiovasc Res, 72: 384-393

[85] Knuefermann P, Schwederski M, Velten M, Krings P, Ehrentraut H, Rudiger M, et al. (2008). Bacterial DNA induces myocardial inflammation and reduces cardiomyocyte contractility: role of toll-like receptor 9. Cardiovasc Res, 78: 26-35

[86] Yin Y, Yan Y, Jiang X, Mai J, Chen NC, Wang H, et al. (2009). Inflammasomes are differentially expressed in cardiovascular and other tissues. Int J Immunopathol Pharmacol, 22: 311-322

[87] Neviere R, Fauvel H, Chopin C, Formstecher P, Marchetti P (2001). Caspase inhibition prevents cardiac dysfunction and heart apoptosis in a rat model of sepsis. Am J Respir Crit Care Med, 163: 218-225

[88] Schmidt C, Kurt B, Hocherl K, Bucher M (2009). Inhibition of NF-kappaB activity prevents downregulation of alpha1-adrenergic receptors and circulatory failure during CLP-induced sepsis. Shock, 32: 239-246

[89] Zang QS, Sadek H, Maass DL, Martinez B, Ma L, Kilgore JA, et al. (2012). Specific inhibition of mitochondrial oxidative stress suppresses inflammation and improves cardiac function in a rat pneumonia-related sepsis model. American journal of physiology. Heart and circulatory physiology, 302: H1847-1859

[90] Oka T, Hikoso S, Yamaguchi O, Taneike M, Takeda T, Tamai T, et al. (2012). Mitochondrial DNA that escapes from autophagy causes inflammation and heart failure. Nature, 485: 251-255

[91] Greenhalgh DG, Saffle JR, Holmes JHt, Gamelli RL, Palmieri TL, Horton JW, et al. (2007). American Burn Association consensus conference to define sepsis and infection in burns. J Burn Care Res, 28: 776-790

[92] Wang L, Quan J, Johnston WE, Maass DL, Horton JW, Thomas JA, et al. (2010). Age-dependent differences of interleukin-6 activity in cardiac function after burn complicated by sepsis. Burns, 36: 232-238

[93] Sheeran PW, Maass DL, White DJ, Turbeville TD, Giroir BP, Horton JW (1998). Aspiration pneumonia-induced sepsis increases cardiac dysfunction after burn trauma. The Journal of surgical research, 76: 192-199

[94] White J, Thomas J, Maass DL, Horton JW (2003). Cardiac effects of burn injury complicated by aspiration pneumonia-induced sepsis. American journal of physiology. Heart and circulatory physiology, 285: H47-58

[95] Tao W, Maass DL, Johnston WE, Horton JW (2005). Murine in vivo myocardial contractile dysfunction after burn injury is exacerbated by pneumonia sepsis. Shock, 24: 495-499

[96] Zang Q, Maass DL, Tsai SJ, Horton JW (2007). Cardiac mitochondrial damage and inflammation responses in sepsis. Surg Infect (Larchmt), 8: 41-54

[97] Reynolds CM, Suliman HB, Hollingsworth JW, Welty-Wolf KE, Carraway MS, Piantadosi CA (2009). Nitric oxide synthase-2 induction optimizes cardiac mitochondrial biogenesis after endotoxemia. Free radical biology & medicine, 46: 564-572

[98] Fauconnier J, Meli AC, Thireau J, Roberge S, Shan J, Sassi Y, et al. Ryanodine receptor leak mediated by caspase-8 activation leads to left ventricular injury after myocardial ischemia-reperfusion. Proceedings of the National Academy of Sciences of the United States of America,

[99] Mizushima N, Levine B (2010). Autophagy in mammalian development and differentiation. Nat Cell Biol, 12: 823-830

[100] Yamamoto S, Sawada K, Shimomura H, Kawamura K, James TN (2000). On the nature of cell death during remodeling of hypertrophied human myocardium. Journal of molecular and cellular cardiology, 32: 161-175

[101] Yan L, Vatner DE, Kim SJ, Ge H, Masurekar M, Massover WH, et al. (2005). Autophagy in chronically ischemic myocardium. Proceedings of the National Academy of Sciences of the United States of America, 102: 13807-13812

[102] Matsui Y, Takagi H, Qu X, Abdellatif M, Sakoda H, Asano T, et al. (2007). Distinct roles of autophagy in the heart during ischemia and reperfusion: roles of AMP-activated protein kinase and Beclin 1 in mediating autophagy. Circulation research, 100: 914-922

[103] Zhu H, Rothermel BA, Hill JA (2009). Autophagy in load-induced heart disease. Methods Enzymol, 453: 343-363

[104] Zhu H, Tannous P, Johnstone JL, Kong Y, Shelton JM, Richardson JA, et al. (2007). Cardiac autophagy is a

maladaptive response to hemodynamic stress. The Journal of clinical investigation, 117: 1782-1793
[105] Liang XH, Jackson S, Seaman M, Brown K, Kempkes B, Hibshoosh H, et al. (1999). Induction of autophagy and inhibition of tumorigenesis by beclin 1. Nature, 402: 672-676
[106] Kroemer G, Marino G, Levine B (2010). Autophagy and the integrated stress response. Mol Cell, 40: 280-293
[107] Levine B, Kroemer G (2008). Autophagy in the pathogenesis of disease. Cell, 132: 27-42
[108] Tannous P, Zhu H, Johnstone JL, Shelton JM, Rajasekaran NS, Benjamin IJ, et al. (2008). Autophagy is an adaptive response in desmin-related cardiomyopathy. Proceedings of the National Academy of Sciences of the United States of America, 105: 9745-9750
[109] Hill JA (2011). Autophagy in cardiac plasticity and disease. Pediatr Cardiol, 32: 282-289
[110] Ferdous A, Battiprolu PK, Ni YG, Rothermel BA, Hill JA (2010). FoxO, autophagy, and cardiac remodeling. Journal of cardiovascular translational research, 3: 355-364
[111] Xie M, Morales CR, Lavandero S, Hill JA (2011). Tuning flux: autophagy as a target of heart disease therapy. Curr Opin Cardiol, 26: 216-222
[112] Inuzuka Y, Okuda J, Kawashima T, Kato T, Niizuma S, Tamaki Y, et al. (2009). Suppression of phosphoinositide 3-kinase prevents cardiac aging in mice. Circulation, 120: 1695-1703
[113] Taneike M, Yamaguchi O, Nakai A, Hikoso S, Takeda T, Mizote I, et al. (2010). Inhibition of autophagy in the heart induces age-related cardiomyopathy. Autophagy, 6: 600-606
[114] Sachs HG, Colgan JA, Lazarus ML (1977). Ultrastructure of the aging myocardium: a morphometric approach. The American journal of anatomy, 150: 63-71
[115] Coleman R, Silbermann M, Gershon D, Reznick AZ (1987). Giant mitochondria in the myocardium of aging and endurance-trained mice. Gerontology, 33: 34-39
[116] Levine B, Mizushima N, Virgin HW (2011). Autophagy in immunity and inflammation. Nature, 469: 323-335
[117] Cuervo AM, Dice JF (2000). When lysosomes get old. Experimental gerontology, 35: 119-131
[118] Nakano M, Oenzil F, Mizuno T, Gotoh S (1995). Age-related changes in the lipofuscin accumulation of brain and heart. Gerontology, 41 Suppl 2: 69-79
[119] Brunk UT, Terman A (2002). Lipofuscin: mechanisms of age-related accumulation and influence on cell function. Free radical biology & medicine, 33: 611-619
[120] Mofarrahi M, Sigala I, Guo Y, Godin R, Davis EC, Petrof B, et al. (2012). Autophagy and skeletal muscles in sepsis. PloS one, 7: e47265
[121] Hsiao HW, Tsai KL, Wang LF, Chen YH, Chiang PC, Chuang SM, et al. (2012). The decline of autophagy contributes to proximal tubular dysfunction during sepsis. Shock, 37: 289-296
[122] Chien WS, Chen YH, Chiang PC, Hsiao HW, Chuang SM, Lue SI, et al. (2011). Suppression of autophagy in rat liver at late stage of polymicrobial sepsis. Shock, 35: 506-511
[123] Watanabe E, Muenzer JT, Hawkins WG, Davis CG, Dixon DJ, McDunn JE, et al. (2009). Sepsis induces extensive autophagic vacuolization in hepatocytes: a clinical and laboratory-based study. Laboratory investigation; a journal of technical methods and pathology, 89: 549-561
[124] Li L, Chen Y, Gibson SB (2013). Starvation-induced autophagy is regulated by mitochondrial reactive oxygen species leading to AMPK activation. Cellular signalling, 25: 50-65
[125] Lee J, Giordano S, Zhang J (2012). Autophagy, mitochondria and oxidative stress: cross-talk and redox signalling. The Biochemical journal, 441: 523-540
[126] Morales CR, Pedrozo Z, Lavandero S, Hill JA (2013). Oxidative Stress and Autophagy in Cardiovascular Homeostasis. Antioxidants & redox signaling,
[127] Hsieh CH, Pai PY, Hsueh HW, Yuan SS, Hsieh YC (2011). Complete induction of autophagy is essential for cardioprotection in sepsis. Annals of surgery, 253: 1190-1200
[128] Yuan H, Perry CN, Huang C, Iwai-Kanai E, Carreira RS, Glembotski CC, et al. (2009). LPS-induced autophagy is mediated by oxidative signaling in cardiomyocytes and is associated with cytoprotection. American journal of physiology. Heart and circulatory physiology, 296: H470-479
[129] Turdi S, Han X, Huff AF, Roe ND, Hu N, Gao F, et al. (2012). Cardiac-specific overexpression of catalase attenuates lipopolysaccharide-induced myocardial contractile dysfunction: role of autophagy. Free radical biology & medicine, 53: 1327-1338
[130] Zang QS, Martinez B, Yao X, Maass DL, Ma L, Wolf SE, et al. (2012). Sepsis-Induced Cardiac Mitochondrial Dysfunction Involves Altered Mitochondrial-Localization of Tyrosine Kinase Src and Tyrosine Phosphatase SHP2. PloS one, 7: e43424
[131] Tibaldi E, Brunati AM, Massimino ML, Stringaro A, Colone M, Agostinelli E, et al. (2008). Src-Tyrosine kinases are major agents in mitochondrial tyrosine phosphorylation. J Cell Biochem, 104: 840-849
[132] Arachiche A, Augereau O, Decossas M, Pertuiset C, Gontier E, Letellier T, et al. (2008). Localization of PTP-1B, SHP-2, and Src exclusively in rat brain mitochondria and functional consequences. The Journal of biological chemistry, 283: 24406-24411
[133] Budas GR, Churchill EN, Disatnik MH, Sun L, Mochly-Rosen D (2010). Mitochondrial import of PKCepsilon is mediated by HSP90: a role in cardioprotection from ischaemia and reperfusion injury. Cardiovascular research, 88: 83-92
[134] Guo J, Cong L, Rybin VO, Gertsberg Z, Steinberg SF (2010). Protein kinase C-{delta} regulates the subcellular localization of Shc in H2O2-treated cardiomyocytes. American journal of physiology. Cell physiology, 299: C770-778
[135] Alonso M, Melani M, Converso D, Jaitovich A, Paz C, Carreras MC, et al. (2004). Mitochondrial extracellular signal-regulated kinases 1/2 (ERK1/2) are modulated during brain development. J Neurochem, 89: 248-256

[136] Monick MM, Powers LS, Barrett CW, Hinde S, Ashare A, Groskreutz DJ, et al. (2008). Constitutive ERK MAPK activity regulates macrophage ATP production and mitochondrial integrity. J Immunol, 180: 7485-7496
[137] Miyazaki T, Neff L, Tanaka S, Horne WC, Baron R (2003). Regulation of cytochrome c oxidase activity by c-Src in osteoclasts. J Cell Biol, 160: 709-718
[138] Feng J, Lucchinetti E, Enkavi G, Wang Y, Gehrig P, Roschitzki B, et al. (2010). Tyrosine phosphorylation by Src within the cavity of the adenine nucleotide translocase 1 regulates ADP/ATP exchange in mitochondria. American journal of physiology. Cell physiology, 298: C740-748
[139] Lewandrowski U, Sickmann A, Cesaro L, Brunati AM, Toninello A, Salvi M (2008). Identification of new tyrosine phosphorylated proteins in rat brain mitochondria. FEBS Lett, 582: 1104-1110
[140] Augereau O, Claverol S, Boudes N, Basurko MJ, Bonneu M, Rossignol R, et al. (2005). Identification of tyrosine-phosphorylated proteins of the mitochondrial oxidative phosphorylation machinery. Cell Mol Life Sci, 62: 1478-1488
[141] Deng N, Zhang J, Zong C, Wang Y, Lu H, Yang P, et al. (2011). Phosphoproteome analysis reveals regulatory sites in major pathways of cardiac mitochondria. Mol Cell Proteomics, 10: M110 000117
[142] Schulenberg B, Aggeler R, Beechem JM, Capaldi RA, Patton WF (2003). Analysis of steady-state protein phosphorylation in mitochondria using a novel fluorescent phosphosensor dye. J Biol Chem, 278: 27251-27255
[143] Chen R, Fearnley IM, Peak-Chew SY, Walker JE (2004). The phosphorylation of subunits of complex I from bovine heart mitochondria. The Journal of biological chemistry, 279: 26036-26045
[144] Steenaart NA, Shore GC (1997). Mitochondrial cytochrome c oxidase subunit IV is phosphorylated by an endogenous kinase. FEBS Lett, 415: 294-298
[145] Ko YH, Pan W, Inoue C, Pedersen PL (2002). Signal transduction to mitochondrial ATP synthase: evidence that PDGF-dependent phosphorylation of the delta-subunit occurs in several cell lines, involves tyrosine, and is modulated by lysophosphatidic acid. Mitochondrion, 1: 339-348
[146] Salvi M, Morrice NA, Brunati AM, Toninello A (2007). Identification of the flavoprotein of succinate dehydrogenase and aconitase as in vitro mitochondrial substrates of Fgr tyrosine kinase. FEBS Lett, 581: 5579-5585
[147] Jakob S, Altschmied J, Haendeler J (2009). "Shping 2" different cellular localizations - a potential new player in aging processes. Aging, 1: 664-668
[148] Chen Q, Vazquez EJ, Moghaddas S, Hoppel CL, Lesnefsky EJ (2003). Production of reactive oxygen species by mitochondria: central role of complex III. The Journal of biological chemistry, 278: 36027-36031
[149] Hebert-Chatelain E, Jose C, Gutierrez Cortes N, Dupuy JW, Rocher C, Dachary-Prigent J, et al. (2012). Preservation of NADH ubiquinone-oxidoreductase activity by Src kinase-mediated phosphorylation of NDUFB10. Biochimica et biophysica acta, 1817: 718-725
[150] Hebert Chatelain E, Dupuy JW, Letellier T, Dachary-Prigent J (2011). Functional impact of PTP1B-mediated Src regulation on oxidative phosphorylation in rat brain mitochondria. Cell Mol Life Sci, 68: 2603-2613
[151] Reznick RM, Shulman GI (2006). The role of AMP-activated protein kinase in mitochondrial biogenesis. The Journal of physiology, 574: 33-39
[152] Reznick RM, Zong H, Li J, Morino K, Moore IK, Yu HJ, et al. (2007). Aging-associated reductions in AMP-activated protein kinase activity and mitochondrial biogenesis. Cell metabolism, 5: 151-156
[153] Nemoto S, Combs CA, French S, Ahn BH, Fergusson MM, Balaban RS, et al. (2006). The mammalian longevity-associated gene product p66shc regulates mitochondrial metabolism. The Journal of biological chemistry, 281: 10555-10560
[154] Cosentino F, Francia P, Camici GG, Pelicci PG, Luscher TF, Volpe M (2008). Final common molecular pathways of aging and cardiovascular disease: role of the p66Shc protein. Arteriosclerosis, thrombosis, and vascular biology, 28: 622-628
[155] Rinaldi S, Landucci F, De Gaudio AR (2009). Antioxidant therapy in critically septic patients. Curr Drug Targets, 10: 872-880
[156] Andrades ME, Ritter C, Dal-Pizzol F (2009). The role of free radicals in sepsis development. Front Biosci (Elite Ed), 1: 277-287
[157] Ritter C, Andrades ME, Reinke A, Menna-Barreto S, Moreira JC, Dal-Pizzol F (2004). Treatment with N-acetylcysteine plus deferoxamine protects rats against oxidative stress and improves survival in sepsis. Critical care medicine, 32: 342-349
[158] Long CL, Maull KI, Krishnan RS, Laws HL, Geiger JW, Borghesi L, et al. (2003). Ascorbic acid dynamics in the seriously ill and injured. The Journal of surgical research, 109: 144-148
[159] Angstwurm MW, Engelmann L, Zimmermann T, Lehmann C, Spes CH, Abel P, et al. (2007). Selenium in Intensive Care (SIC): results of a prospective randomized, placebo-controlled, multiple-center study in patients with severe systemic inflammatory response syndrome, sepsis, and septic shock. Crit Care Med, 35: 118-126
[160] Forceville X, Laviolle B, Annane D, Vitoux D, Bleichner G, Korach JM, et al. (2007). Effects of high doses of selenium, as sodium selenite, in septic shock: a placebo-controlled, randomized, double-blind, phase II study. Crit Care, 11: R73
[161] Berger MM, Chiolero RL (2007). Antioxidant supplementation in sepsis and systemic inflammatory response syndrome. Critical care medicine, 35: S584-590
[162] Lovat R, Preiser JC (2003). Antioxidant therapy in intensive care. Curr Opin Crit Care, 9: 266-270
[163] Adlam VJ, Harrison JC, Porteous CM, James AM, Smith RA, Murphy MP, et al. (2005). Targeting an antioxidant to mitochondria decreases cardiac ischemia-reperfusion injury. FASEB journal : official publication of the

Federation of American Societies for Experimental Biology, 19: 1088-1095
[164] Szeto HH (2006). Cell-permeable, mitochondrial-targeted, peptide antioxidants. AAPS J, 8: E277-283
[165] Amstislavskaya TG, Maslova LN, Gladkikh DV, Belousova, II, Stefanova NA, Kolosova NG Effects of the mitochondria-targeted antioxidant SkQ1 on sexually motivated behavior in male rats. Pharmacol Biochem Behav,
[166] Demianenko IA, Vasilieva TV, Domnina LV, Dugina VB, Egorov MV, Ivanova OY, et al. Novel mitochondria-targeted antioxidants, "Skulachev-ion" derivatives, accelerate dermal wound healing in animals. Biochemistry. Biokhimiia, 75: 274-280
[167] Coulter CV, Kelso GF, Lin TK, Smith RA, Murphy MP (2000). Mitochondrially targeted antioxidants and thiol reagents. Free radical biology & medicine, 28: 1547-1554
[168] Murphy MP, Smith RA (2007). Targeting antioxidants to mitochondria by conjugation to lipophilic cations. Annual review of pharmacology and toxicology, 47: 629-656
[169] Smith RA, Porteous CM, Gane AM, Murphy MP (2003). Delivery of bioactive molecules to mitochondria in vivo. Proceedings of the National Academy of Sciences of the United States of America, 100: 5407-5412
[170] Szeto HH (2008). Mitochondria-targeted cytoprotective peptides for ischemia-reperfusion injury. Antioxidants & redox signaling, 10: 601-619
[171] Lowes DA, Wallace C, Murphy MP, Webster NR, Galley HF (2009). The mitochondria targeted antioxidant MitoQ protects against fluoroquinolone-induced oxidative stress and mitochondrial membrane damage in human Achilles tendon cells. Free radical research, 43: 323-328
[172] Dhanasekaran A, Kotamraju S, Kalivendi SV, Matsunaga T, Shang T, Keszler A, et al. (2004). Supplementation of endothelial cells with mitochondria-targeted antioxidants inhibit peroxide-induced mitochondrial iron uptake, oxidative damage, and apoptosis. J Biol Chem, 279: 37575-37587
[173] Dhanasekaran A, Kotamraju S, Karunakaran C, Kalivendi SV, Thomas S, Joseph J, et al. (2005). Mitochondria superoxide dismutase mimetic inhibits peroxide-induced oxidative damage and apoptosis: role of mitochondrial superoxide. Free Radic Biol Med, 39: 567-583
[174] Jauslin ML, Meier T, Smith RA, Murphy MP (2003). Mitochondria-targeted antioxidants protect Friedreich Ataxia fibroblasts from endogenous oxidative stress more effectively than untargeted antioxidants. FASEB J, 17: 1972-1974
[175] Gane EJ, Weilert F, Orr DW, Keogh GF, Gibson M, Lockhart MM, et al. (2010). The mitochondria-targeted anti-oxidant mitoquinone decreases liver damage in a phase II study of hepatitis C patients. Liver international : official journal of the International Association for the Study of the Liver, 30: 1019-1026
[176] Smith RA, Murphy MP (2010). Animal and human studies with the mitochondria-targeted antioxidant MitoQ. Annals of the New York Academy of Sciences, 1201: 96-103
[177] Ghosh A, Chandran K, Kalivendi SV, Joseph J, Antholine WE, Hillard CJ, et al. (2010). Neuroprotection by a mitochondria-targeted drug in a Parkinson's disease model. Free radical biology & medicine, 49: 1674-1684
[178] Manczak M, Mao P, Calkins MJ, Cornea A, Reddy AP, Murphy MP, et al. (2010). Mitochondria-targeted antioxidants protect against amyloid-beta toxicity in Alzheimer's disease neurons. Journal of Alzheimer's disease : JAD, 20 Suppl 2: S609-631
[179] Chandran K, Aggarwal D, Migrino RQ, Joseph J, McAllister D, Konorev EA, et al. (2009). Doxorubicin inactivates myocardial cytochrome c oxidase in rats: cardioprotection by Mito-Q. Biophys J, 96: 1388-1398
[180] Dikalova AE, Bikineyeva AT, Budzyn K, Nazarewicz RR, McCann L, Lewis W, et al. Therapeutic targeting of mitochondrial superoxide in hypertension. Circulation research, 107: 106-116
[181] Chacko BK, Reily C, Srivastava A, Johnson MS, Ye Y, Ulasova E, et al. (2010). Prevention of diabetic nephropathy in Ins2(+/)(AkitaJ) mice by the mitochondria-targeted therapy MitoQ. The Biochemical journal, 432: 9-19
[182] Lowes DA, Thottakam BM, Webster NR, Murphy MP, Galley HF (2008). The mitochondria-targeted antioxidant MitoQ protects against organ damage in a lipopolysaccharide-peptidoglycan model of sepsis. Free radical biology & medicine, 45: 1559-1565
[183] Supinski GS, Murphy MP, Callahan LA (2009). MitoQ administration prevents endotoxin-induced cardiac dysfunction. Am J Physiol Regul Integr Comp Physiol, 297: R1095-1102
[184] Antonenko YN, Avetisyan AV, Bakeeva LE, Chernyak BV, Chertkov VA, Domnina LV, et al. (2008). Mitochondria-targeted plastoquinone derivatives as tools to interrupt execution of the aging program. 1. Cationic plastoquinone derivatives: synthesis and in vitro studies. Biochemistry. Biokhimiia, 73: 1273-1287
[185] Stefanova NA, Fursova A, Kolosova NG (2010). Behavioral effects induced by mitochondria-targeted antioxidant SkQ1 in Wistar and senescence-accelerated OXYS rats. Journal of Alzheimer's disease : JAD, 21: 479-491
[186] Shipounova IN, Svinareva DA, Petrova TV, Lyamzaev KG, Chernyak BV, Drize NI, et al. (2010). Reactive oxygen species produced in mitochondria are involved in age-dependent changes of hematopoietic and mesenchymal progenitor cells in mice. A study with the novel mitochondria-targeted antioxidant SkQ1. Mechanisms of ageing and development, 131: 415-421
[187] Krementsova AV, Roshina NV, Tsybul'ko EA, Rybina OY, Symonenko AV, Pasyukova EG (2012). Reproducible effects of the mitochondria-targeted plastoquinone derivative SkQ1 on Drosophila melanogaster lifespan under different experimental scenarios. Biogerontology, 13: 595-607
[188] Anisimov VN, Egorov MV, Krasilshchikova MS, Lyamzaev KG, Manskikh VN, Moshkin MP, et al.

(2011). Effects of the mitochondria-targeted antioxidant SkQ1 on lifespan of rodents. Aging, 3: 1110-1119

[189] Venkataraman R, Subramanian S, Kellum JA (2003). Clinical review: extracorporeal blood purification in severe sepsis. Crit Care, 7: 139-145