α-Lipoic Acid and Vitamin C in Isoprenaline-Induced Myocardial Injury: Physiological Mechanisms and Cardioprotective Potential

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Abstract

Background:: Isoprenaline-induced myocardial injury is a reproducible experimental model used to investigate the biochemical and cellular consequences of excessive β-adrenergic stimulation. At high doses, isoprenaline markedly increases cardiac workload and undergoes auto-oxidation, initiating reactive oxygen species generation, lipid peroxidation, calcium dysregulation, mitochondrial impairment, inflammation, and cardiomyocyte death. These interconnected abnormalities resemble important components of acute myocardial injury, although the model does not reproduce coronary atherothrombosis in humans.

Objective: This review examines the physiological basis of isoprenaline-induced myocardial injury and evaluates the potential cardioprotective actions of α-lipoic acid and vitamin C, individually and in combination.

Main body: α-Lipoic acid is a mitochondrial organosulfur compound with metabolic, antioxidant, and signaling functions. Its reduced form, dihydrolipoic acid, participates in a redox couple capable of scavenging reactive species, chelating redox-active metals, regenerating endogenous antioxidants, and supporting glutathione homeostasis. α-Lipoic acid may also preserve mitochondrial bioenergetics and suppress redox-sensitive inflammatory and apoptotic pathways. Vitamin C is a water-soluble electron donor that neutralizes several reactive species, limits lipid and protein oxidation, regenerates other antioxidants, and contributes to endothelial and inflammatory regulation. Experimental evidence indicates that vitamin C can attenuate cardiac biomarker release, oxidative injury, electrocardiographic abnormalities, and histopathological damage following isoprenaline exposure. The different physicochemical properties of α-lipoic acid and vitamin C provide a plausible basis for complementary protection across aqueous, membranous, and mitochondrial cellular compartments. Nevertheless, direct evidence establishing synergistic cardioprotection remains limited.

Conclusion: Oxidative stress is a central component of isoprenaline cardiotoxicity but operates within a wider network involving calcium overload, mitochondrial failure, inflammation, and regulated cell death. α-Lipoic acid and vitamin C may interrupt several components of this network. Their combined administration is physiologically rational, although carefully designed experimental studies are required to distinguish additive effects from genuine pharmacological synergy and to determine their translational relevance.

Keywords: α-Lipoic acid, Ascorbic acid, Isoprenaline, Myocardial injury, Oxidative stress

Introduction

Acute myocardial injury is the final expression of interacting metabolic, ionic, mitochondrial, inflammatory, and cell-death processes. In human myocardial infarction, prolonged ischemia restricts oxygen delivery, rapidly reduces oxidative phosphorylation, depletes adenosine triphosphate, and disturbs membrane ion transport. Intracellular sodium and calcium subsequently accumulate, mitochondrial function deteriorates, and reactive oxygen species amplify damage to proteins, lipids, and nucleic acids. Although restoration of blood flow is essential for myocardial salvage, reperfusion itself may produce further oxidative and calcium-mediated injury. Experimental models remain indispensable for dissecting these mechanisms and evaluating interventions before clinical translation.[1]

Among the available preclinical approaches, high-dose isoprenaline administration offers a technically simple and reproducible model of diffuse myocardial injury without surgical coronary ligation.

Isoprenaline, also known as isoproterenol, is a synthetic nonselective β-adrenergic agonist. Excessive stimulation of β_1-adrenergic receptors increases heart rate, myocardial contractility, and oxygen consumption, creating a pronounced imbalance between cardiac oxygen demand and supply. Simultaneously, oxidation of isoprenaline generates reactive intermediates and promotes oxidative deterioration of cardiomyocyte membranes. The resulting lesions are therefore produced predominantly by catecholamine overload, metabolic stress, and direct cardiotoxicity rather than by coronary atherothrombosis.[2,3]

The injury evolves through a closely interconnected sequence. β-Adrenergic overstimulation increases cyclic adenosine monophosphate signaling and calcium entry, while energy depletion limits the capacity of cardiomyocytes to restore ionic homeostasis. Cytosolic and mitochondrial calcium accumulation promotes contractile dysfunction, protease activation, mitochondrial membrane destabilization, and further reactive oxygen species generation. Lipid peroxidation compromises sarcolemmal integrity, permitting leakage of cardiac enzymes, whereas activation of nuclear factor-κB and other redox-sensitive pathways stimulates inflammatory cytokine production. When these disturbances exceed cellular adaptive capacity, cardiomyocytes undergo necrotic and apoptotic death, accompanied by characteristic electrocardiographic, biochemical, and histopathological abnormalities.[3-5]

This strong oxidative component makes the isoprenaline model particularly suitable for studying redox-directed cardioprotection. α-Lipoic acid is an endogenous disulfide-containing compound that functions as a cofactor for mitochondrial enzyme complexes involved in oxidative metabolism. Together with its reduced form, dihydrolipoic acid, it forms a versatile redox system with activity in both aqueous and lipid-associated environments. Beyond direct radical neutralization, α-lipoic acid may chelate redox-active metals, regenerate glutathione and vitamins C and E, improve endogenous antioxidant capacity, and regulate inflammatory and apoptotic signaling. These properties suggest that its cardioprotective value may extend from cytosolic antioxidant defense to preservation of mitochondrial metabolism and cellular survival.[6]

Vitamin C, or ascorbic acid, is a major water-soluble antioxidant and an essential enzymatic cofactor. By donating electrons, it can neutralize reactive oxygen and nitrogen species, reduce oxidized biomolecules, and regenerate vitamin E within biological membranes. Vitamin C also influences endothelial nitric oxide availability, leukocyte activity, cytokine signaling, and cellular redox regulation. In isoprenaline-treated rats, ascorbic acid administration has reduced biochemical and histological evidence of myocardial damage, supporting its capacity to counter catecholamine-associated oxidative cardiotoxicity.[7,8]

Because α-lipoic acid can operate across mitochondrial, lipid, and aqueous environments and can participate in the recycling of vitamin C, combined administration may provide broader redox coverage than either compound alone. Such an interaction could theoretically preserve antioxidant reserves, restrict membrane lipid peroxidation, stabilize mitochondrial function, and attenuate downstream inflammatory and cell-death pathways. This proposed complementarity remains mechanistically attractive but should not be described as synergy until combined treatment is shown to produce an effect exceeding the expected actions of the individual agents.[6-8]

Accordingly, this review examines the physiological mechanisms responsible for isoprenaline-induced myocardial injury and critically evaluates the cardioprotective potential of α-lipoic acid and vitamin C. Particular attention is given to β-adrenergic overstimulation, oxidative stress, calcium dysregulation, mitochondrial dysfunction, inflammation, apoptosis, and the biological rationale for combining these antioxidants

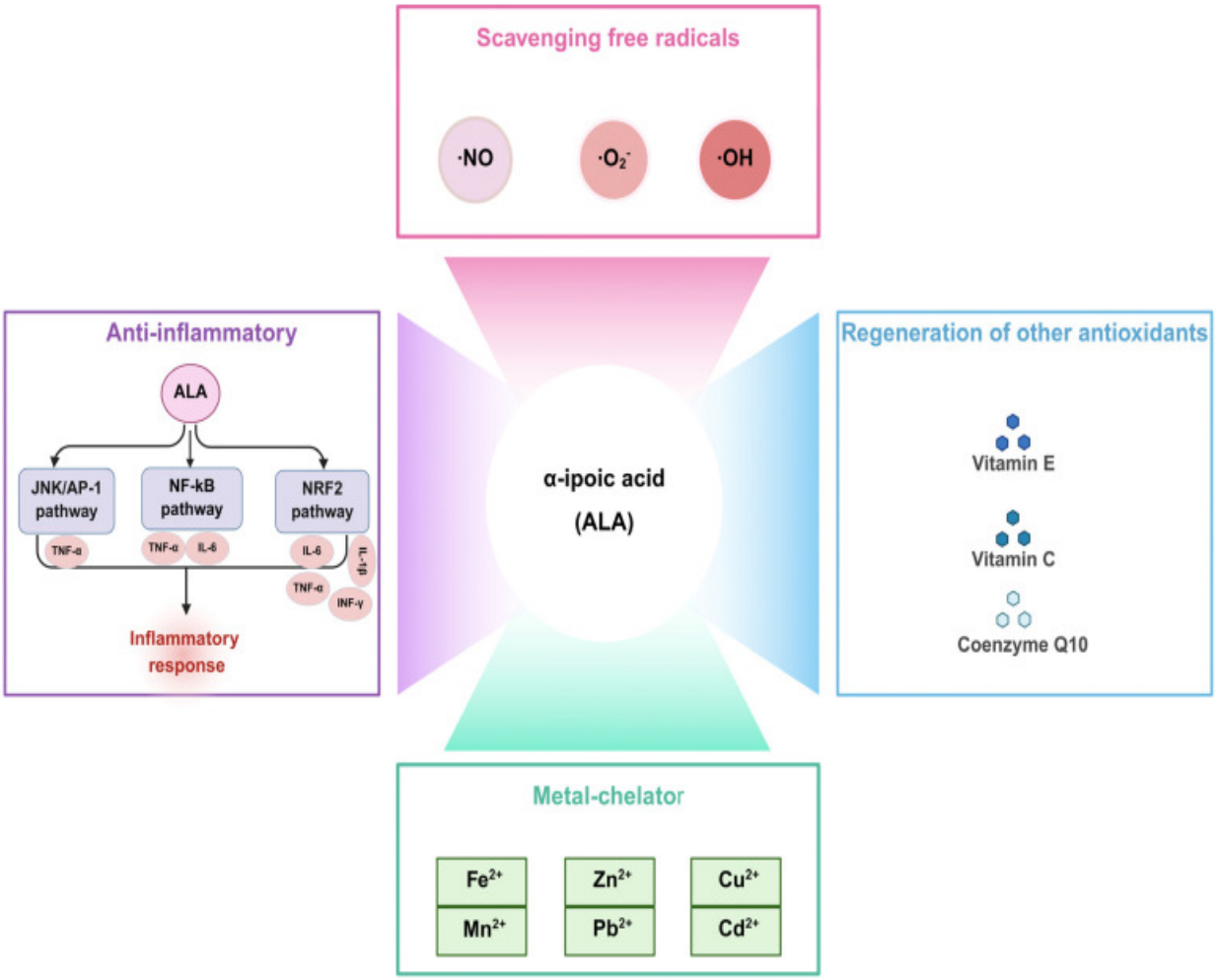

Figure 1: Multiple biological functions of ALA. ALA exerts antioxidant effects by scavenging reactive species, including •NO, •O_2^-, and •OH. It also inhibits inflammation by modulating the JNK/AP-1, NF-κB, and NRF2 pathways, which reduce the expression of inflammatory factors, such as TNF-α, IL-6, IL-1β, and INF-γ. Additionally, ALA can regenerate other antioxidants, including vitamin C, vitamin E, and coenzyme Q10. As a metal chelator, ALA binds to metal ions (Fe^{2+}, Zn^{2+}, Cu^{2+}, Mn^{2+}, Pb^{2+}, and Cd^{2+}).(**Wang Y,et al 2025). ALA**, α-lipoic acid; **NO**, nitric oxide; **O_2•$^-$**, superoxide anion; **•OH**, hydroxyl radical; **JNK/AP-1, c-Jun N**-terminal kinase/activator protein-1**; NF-κB**, nuclear factor kappa B; **Nrf2**, nuclear factor erythroid 2-related factor 2; **TNF-α**, tumor necrosis factor-alpha; IL-6, interleukin-6; **IL-1β**, interleukin-1 beta; **IFN**-γ, interferon-gamma. [6]

Isoprenaline-Induced Myocardial Injury as an Experimental Model

Isoprenaline is a synthetic catecholamine with nonselective agonistic activity at β_1- and β_2-adrenergic receptors. Excessive β_1-receptor stimulation increases cyclic adenosine monophosphate production, protein kinase A activity, calcium entry, heart rate, and myocardial contractility. These changes markedly elevate cardiac oxygen and substrate requirements. Meanwhile, β_2-mediated peripheral vasodilatation may reduce diastolic blood pressure and compromise coronary perfusion during heightened myocardial demand, aggravating the imbalance between oxygen supply and consumption.[2]

In addition to its hemodynamic effects, isoprenaline causes direct chemical injury to cardiomyocytes. Oxidation of its catechol structure generates quinone derivatives, semiquinone radicals, superoxide, and hydrogen peroxide. These reactive intermediates promote membrane lipid peroxidation, oxidize protein sulfhydryl groups, and interfere with enzymes responsible for cellular metabolism and ion regulation. The model therefore combines intense β-adrenergic stimulation with catecholamine-derived oxidative toxicity.[3]

The extent of myocardial injury varies according to the administered dose, route of delivery, dosing schedule, animal species and strain, age, sex, and timing of tissue assessment. Acute protocols commonly employ one or two large subcutaneous or intraperitoneal doses administered approximately 24 hours apart. Such regimens predominantly produce myocardial necrosis and acute inflammation, whereas repeated lower-dose exposure may induce progressive ventricular hypertrophy, fibrosis, and functional deterioration.[10]

Histopathological examination typically demonstrates myofibrillar disorganization, contraction-band lesions, cytoplasmic vacuolation, interstitial edema, inflammatory-cell infiltration, and focal or confluent cardiomyocyte necrosis. Loss of sarcolemmal integrity permits cardiac troponins and intracellular enzymes to enter the circulation. These changes may be accompanied by electrocardiographic abnormalities, including ST-segment displacement, altered R-wave amplitude, pathological Q waves, tachycardia, and rhythm disturbances.[11]

Although this experimental condition is frequently termed isoprenaline-induced myocardial infarction, that description should be interpreted cautiously. The model reproduces several important consequences of acute myocardial damage, including oxidative stress, calcium overload, mitochondrial dysfunction, inflammation, biomarker release, and cardiomyocyte death. However, it does not reproduce plaque rupture, coronary thrombosis, persistent regional arterial occlusion, or the anatomically defined ischemic territory characteristic of human atherothrombotic myocardial infarction.[1]

Physiological Mechanisms of Isoprenaline-Induced Myocardial Injury

β-Adrenergic Overstimulation and Oxygen Imbalance

Sustained β_1-adrenergic activation increases adenylyl cyclase activity, cyclic adenosine monophosphate formation, and protein kinase A–dependent phosphorylation of L-type calcium channels and calcium-handling proteins. The resulting increases in chronotropy and inotropy substantially elevate myocardial oxygen consumption. When this demand cannot be matched by coronary oxygen delivery, cardiomyocytes develop relative hypoxia, impaired oxidative phosphorylation, and progressive depletion of adenosine triphosphate.[2]

The rise in heart rate also shortens diastole, the principal period of coronary perfusion, while β_2-mediated systemic vasodilatation may reduce diastolic arterial pressure. These simultaneous changes intensify the mismatch between myocardial workload and oxygen availability. Subendocardial regions are particularly vulnerable because they experience greater wall stress and receive proportionally less perfusion when diastole is shortened.[10]

Oxidative Stress and Lipid Peroxidation

The oxidative transformation of isoprenaline generates reactive intermediates capable of transferring electrons to molecular oxygen. Superoxide may subsequently form hydrogen peroxide and, in the presence of redox-active iron, the highly reactive hydroxyl radical. Reactive oxygen species also arise from mitochondrial electron leakage, xanthine oxidase activity, activated inflammatory cells, and uncoupled enzymatic reactions, creating several mutually reinforcing sources of oxidative stress.[3]

Polyunsaturated fatty acids within the sarcolemma and intracellular membranes are major targets of free-radical attack. Lipid peroxidation decreases membrane fluidity, alters ion-channel function, disrupts membrane-bound enzymes, and generates reactive aldehydes that propagate injury beyond the initial oxidation site. Increasing malondialdehyde and lipid hydroperoxide concentrations, accompanied by diminished myocardial superoxide dismutase, catalase, and glutathione activity, are therefore commonly observed in isoprenaline-treated animals.[9]

Oxidative injury also affects proteins and nucleic acids. Oxidation of sulfhydryl groups can alter calcium pumps, metabolic enzymes, contractile proteins, and components of the mitochondrial respiratory chain. DNA oxidation activates repair pathways that consume cellular energy, while extensive molecular damage overwhelms antioxidant defenses and promotes cardiomyocyte dysfunction and death.[5]

Calcium Dysregulation

Calcium overload is a central link between β-adrenergic stimulation and structural myocardial damage. Protein kinase A–dependent enhancement of L-type calcium-channel activity increases calcium entry during each action potential, while accelerated sarcoplasmic reticulum cycling enlarges the intracellular calcium burden. When energy availability declines and membrane integrity deteriorates, calcium extrusion through the sarcolemmal calcium pump and sodium-calcium exchanger becomes progressively ineffective.[4]

Excess cytosolic calcium activates calpains, phospholipases, endonucleases, and other calcium-dependent enzymes that degrade cytoskeletal and membrane components. Mitochondrial calcium uptake initially buffers the cytosolic rise but eventually promotes permeability-transition pore opening, membrane depolarization, respiratory failure, and further reactive oxygen species generation. Oxidative stress and calcium overload consequently form a self-amplifying cycle rather than independent pathways.[4]

Mitochondrial Dysfunction and Energy Failure

Mitochondria sustain cardiac contraction by continuously generating adenosine triphosphate through oxidative phosphorylation. Isoprenaline-induced oxygen imbalance, calcium accumulation, and oxidative modification of respiratory-chain proteins reduce mitochondrial efficiency. Electron leakage then increases, mitochondrial membrane potential declines, and adenosine triphosphate production becomes inadequate for contraction, ion transport, and cellular repair.[12]

Severe mitochondrial damage determines whether the cardiomyocyte remains viable. Permeability-transition pore opening causes matrix swelling, loss of membrane potential, and interruption of oxidative phosphorylation. Rupture or permeabilization of the outer mitochondrial membrane releases cytochrome c and other proapoptotic proteins, whereas profound energy depletion favors membrane failure and necrotic death.[12]

Inflammation and Redox-Sensitive Signaling

Cellular injury activates nuclear factor-κB and other redox-sensitive transcriptional pathways. These signals increase the expression of tumor necrosis factor-α, interleukin-1β, interleukin-6, adhesion molecules, and chemotactic mediators. Recruitment of neutrophils and monocytes assists in removing damaged tissue but also expands myocardial injury through protease release, oxidative bursts, and additional cytokine production.[5]

Damage-associated molecular patterns released from necrotic cardiomyocytes can activate pattern-recognition receptors and inflammasome signaling. Assembly of the nucleotide-binding oligomerization domain-like receptor protein 3 inflammasome promotes caspase-1 activation and maturation of interleukin-1β and interleukin-18. Although these mechanisms have been studied most extensively in ischemic infarction, they provide a relevant framework for understanding inflammatory amplification after chemically induced cardiomyocyte necrosis.[13]

Apoptosis, Necrosis, and Subsequent Remodeling

Moderate mitochondrial injury may initiate intrinsic apoptosis through cytochrome c release, apoptosome formation, and sequential activation of caspase-9 and caspase-3. An increased Bax-to-Bcl-2 ratio favors mitochondrial permeabilization, whereas severe oxidative damage and adenosine triphosphate depletion produce uncontrolled membrane rupture and necrosis. Both patterns may coexist within isoprenaline-injured myocardium, with their relative contribution varying according to dose and assessment time.[5]

Following acute injury, inflammatory cells remove necrotic tissue and fibroblasts deposit extracellular matrix to preserve mechanical integrity. Excessive or prolonged collagen deposition reduces ventricular

compliance, interferes with electrical conduction, and contributes to maladaptive remodeling. Repeated or extended isoprenaline exposure can therefore progress from acute cardiomyocyte injury to ventricular hypertrophy, fibrosis, and persistent functional impairment.[10]

Assessment of Myocardial Injury in the Isoprenaline Model

Serum cardiac troponin I or T provides the most cardiac-specific biochemical evidence of cardiomyocyte injury. Creatine kinase-MB and lactate dehydrogenase may also increase because of sarcolemmal disruption, although they are less specific for myocardial tissue. Experimental interpretation should integrate circulating biomarkers with histopathology because biomarker release alone does not identify the mechanism, distribution, or morphological severity of injury.[1]

Oxidative status is commonly evaluated by measuring malondialdehyde, thiobarbituric acid-reactive substances, lipid hydroperoxides, protein carbonyls, or reduced glutathione. Activities of superoxide dismutase, catalase, glutathione peroxidase, and glutathione reductase provide complementary information regarding endogenous antioxidant capacity. No individual assay completely characterizes redox balance; consequently, a panel containing damage markers and antioxidant measures is preferable to reliance on one endpoint.[14]

Electrocardiographic assessment may demonstrate changes in heart rate, ST segment, QRS complex, R-wave amplitude, QT interval, or rhythm. Histopathological evaluation can grade myofibrillar degeneration, edema, inflammatory infiltration, vascular congestion, contraction-band injury, and necrosis. Blinded scoring and prespecified criteria improve reliability, particularly when lesions are multifocal and tissue sampling may influence the observed severity.[11]

α-Lipoic Acid: Physiological and Pharmacological Basis

Biochemistry and Redox Properties

α-Lipoic acid is an eight-carbon organosulfur compound containing a cyclic disulfide group. Endogenously synthesized lipoic acid is covalently attached to lysine residues within mitochondrial enzyme complexes, including pyruvate dehydrogenase and α-ketoglutarate dehydrogenase. In this protein-bound form, it participates in oxidative decarboxylation and links substrate metabolism to mitochondrial energy generation.[6]

Exogenously administered α-lipoic acid differs functionally from enzyme-bound lipoate. It is absorbed, distributed to tissues, and reduced intracellularly to dihydrolipoic acid. The α-lipoic acid–dihydrolipoic acid couple can participate in electron-transfer reactions, reduce selected oxidized biomolecules, and influence the cellular thiol-redox environment. Its actions therefore extend beyond simple direct scavenging of free radicals.[15]

Reinforcement of Antioxidant Defenses

α-Lipoic acid may support glutathione homeostasis by increasing cysteine availability and facilitating the reduction of oxidized glutathione. It can also contribute indirectly to the regeneration of vitamin C and vitamin E and may chelate certain redox-active metal ions. These properties allow antioxidant protection to operate across aqueous, membranous, and mitochondrial compartments.[15]

Another important action involves activation of nuclear factor erythroid 2-related factor 2. Following its release from Kelch-like ECH-associated protein 1, nuclear factor erythroid 2-related factor 2 translocates to the nucleus and promotes transcription of antioxidant and cytoprotective enzymes. These include heme oxygenase-1, glutathione-related enzymes, and nicotinamide adenine dinucleotide phosphate-generating systems that strengthen endogenous resistance to oxidative stress.[6]

Mitochondrial Protection

Preservation of mitochondrial function may be especially relevant to the isoprenaline model because mitochondrial calcium overload and respiratory impairment are major determinants of cardiomyocyte survival. α-Lipoic acid has been associated experimentally with reduced mitochondrial reactive oxygen species formation, maintenance of membrane potential, improved respiratory activity, and limitation of permeability-transition pore opening.[16]

In myocardial ischemia-reperfusion experiments, α-lipoic acid pretreatment reduced infarct injury and

improved mitochondrial aldehyde dehydrogenase-2 activity. This enzyme detoxifies reactive aldehydes produced during lipid peroxidation, providing a plausible connection between α-lipoic acid, preservation of mitochondrial function, and reduced propagation of oxidative membrane damage.[16]

Anti-Inflammatory and Antiapoptotic Actions

By reducing oxidative activation of nuclear factor-κB, α-lipoic acid may decrease the production of inflammatory cytokines and adhesion molecules. It may also limit proapoptotic signaling by reducing the Bax-to-Bcl-2 ratio, preventing cytochrome c release, and suppressing downstream caspase activation. These effects could protect cardiomyocytes even when direct radical scavenging alone is insufficient to control injury.[6]

Experimental studies of myocardial ischemia-reperfusion support the ability of α-lipoic acid to reduce infarct size and preserve ventricular performance under selected conditions. However, these models involve coronary occlusion and reperfusion rather than isoprenaline toxicity, and their findings cannot be transferred uncritically. Direct investigation within the isoprenaline model remains necessary to confirm the effective dose, timing, and dominant protective pathway.[17]

Pharmacokinetic Considerations and Limitations

Oral α-lipoic acid undergoes rapid absorption but has limited and variable bioavailability because of presystemic metabolism, competition with food, and chemical instability. Both R- and S-enantiomers may be present in commercial preparations, although naturally occurring enzyme-bound lipoic acid is the R-form. Experimental studies should therefore report the formulation, enantiomeric composition, administration route, dose, and timing in relation to isoprenaline exposure.[6]

The redox activity of α-lipoic acid is context-dependent. At inappropriate concentrations or under particular metal and oxygen conditions, redox-active molecules may display pro-oxidant behavior. Furthermore, improvement in isolated antioxidant markers does not necessarily demonstrate meaningful cardioprotection. Evidence should include structural or functional cardiac outcomes in addition to biochemical measurements.[15]

Vitamin C: Physiological and Cardioprotective Actions

Redox Biology of Vitamin C

Vitamin C is a water-soluble electron donor that exists principally as ascorbate at physiological pH. Humans require dietary vitamin C because they lack functional gulonolactone oxidase, whereas rats synthesize ascorbate endogenously. This species difference is important when interpreting supplementation studies because experimentally administered vitamin C increases an already existing endogenous pool in rats rather than correcting an obligatory dietary deficiency.[7]

Ascorbate can neutralize superoxide-derived species, aqueous peroxyl radicals, hypochlorous acid, and several reactive nitrogen intermediates. Following electron donation, the relatively stable ascorbyl radical is generated and can be recycled to ascorbate. If further oxidized to dehydroascorbic acid, it may be reduced through glutathione- and enzyme-dependent systems or irreversibly degraded when recycling capacity is exceeded.[7]

Vitamin C also regenerates α-tocopherol from the tocopheroxyl radical at the membrane-aqueous interface. This interaction is physiologically important because vitamin E interrupts lipid-peroxidation chain reactions within membranes, while vitamin C restores its reduced antioxidant form from the surrounding aqueous phase. Vitamin C consequently supports membrane protection despite being primarily water soluble.[18]

Effects on Inflammation and Vascular Function

Ascorbate can influence inflammatory responses by limiting oxidant-mediated activation of nuclear factor-κB, supporting leukocyte redox regulation, and reducing oxidative damage generated during the respiratory burst. Its actions are not uniformly immunosuppressive; rather, adequate ascorbate availability may support normal host-defense functions while restricting collateral oxidative injury.[18]

Vitamin C may preserve endothelial nitric oxide signaling by limiting oxidation of tetrahydrobiopterin, a cofactor required for coupled endothelial nitric oxide synthase activity. When tetrahydrobiopterin is

oxidized, the enzyme may generate superoxide rather than nitric oxide. Preservation of nitric oxide availability can support vasodilatation and microvascular perfusion, although the importance of this mechanism in high-dose isoprenaline injury requires direct confirmation.[19]

Evidence in Isoprenaline-Induced Myocardial Injury

In rats exposed to isoprenaline, vitamin C treatment has been associated with lower cardiac biomarker release, reduced lipid peroxidation, preservation of endogenous antioxidant activity, and less severe histological injury. These observations indicate that ascorbate can attenuate important manifestations of catecholamine-induced cardiotoxicity rather than merely altering a single biochemical endpoint.[8]

Vitamin C has also reduced electrocardiographic abnormalities and structural myocardial damage in experimental isoprenaline injury. The findings are consistent with membrane stabilization and diminished oxidative injury, although the extent of protection depends on the dose, pretreatment period, route of administration, and severity of the isoprenaline protocol.[20]

Limitations of Vitamin C Cardioprotection

Vitamin C absorption is saturable, tissue distribution is tightly regulated, and excess circulating ascorbate is eliminated through the kidneys. Increasing the oral dose therefore does not produce a proportional increase in plasma concentration. Experimental comparisons must distinguish oral supplementation from parenteral administration because the latter can achieve substantially higher circulating concentrations and may have different biological effects.[19]

Ascorbate may also act as a pro-oxidant in vitro when it reduces free transition metals that subsequently participate in radical-generating reactions. Under physiological conditions, iron and copper are generally protein-bound, making this concern less straightforward in vivo. Nevertheless, dose selection should be biologically justified, and antioxidant benefit should be demonstrated through convergent biochemical, histological, and functional outcomes.[7]

Rationale for Combining α-Lipoic Acid and Vitamin C

The combination is attractive because the two compounds differ in distribution and mechanism. Vitamin C acts predominantly in aqueous environments and at membrane interfaces, whereas α-lipoic acid and dihydrolipoic acid can influence mitochondrial metabolism, thiol balance, antioxidant recycling, and redox-sensitive signaling. Their overlapping but nonidentical activities could provide broader cellular protection than either agent alone.[15]

Dihydrolipoic acid can reduce dehydroascorbic acid under experimental conditions, potentially assisting the regeneration of vitamin C. Recycled ascorbate can then support the restoration of vitamin E and restrict membrane lipid peroxidation. This interconnected antioxidant network provides a plausible physiological basis for combined therapy, especially when isoprenaline simultaneously affects mitochondria, cytosolic redox balance, and membrane integrity.[15]

Potential complementarity may also extend beyond antioxidant recycling. α-Lipoic acid could preserve mitochondrial bioenergetics and suppress inflammatory or apoptotic signaling, while vitamin C could strengthen aqueous antioxidant defense and support endothelial nitric oxide availability. Coordinated effects at these different sites might reduce cardiac enzyme leakage, lipid peroxidation, inflammatory infiltration, and cardiomyocyte death.[6]

However, mechanistic plausibility is not proof of synergy. A combined regimen is synergistic only when its effect exceeds the expected combined effects of the separate treatments. A study containing only untreated, isoprenaline, and combined-treatment groups cannot determine whether the interaction is additive, synergistic, or driven by one compound.[21]

A rigorous experiment should therefore include healthy control, isoprenaline control, α-lipoic acid alone, vitamin C alone, and combined α-lipoic acid plus vitamin C groups. Equivalent exposure periods and appropriate vehicle controls are essential. Analysis should formally test the interaction between the two interventions rather than infer synergy from a larger numerical improvement in the combined group.[21]

Experimental Outcomes Needed to Establish Cardioprotection

Evaluation should encompass several complementary physiological domains. Circulating cardiac

troponin and creatine kinase-MB can indicate membrane injury, whereas malondialdehyde, reduced glutathione, superoxide dismutase, catalase, and glutathione peroxidase can characterize redox balance. Proinflammatory cytokines, nuclear factor-κB activity, Bax, Bcl-2, and caspase-3 can clarify inflammatory and apoptotic mechanisms.[5]

Histopathological examination remains necessary to confirm whether biomarker improvement corresponds to preserved myocardial structure. Blinded assessment should document myofibrillar degeneration, edema, inflammatory infiltration, hemorrhage, vascular congestion, and necrosis. Where available, echocardiographic indices, hemodynamic measurements, or ex vivo cardiac performance would provide stronger evidence that biochemical protection produces functional benefit.[1]

Assessment timing must match the biological question. Early sampling emphasizes oxidative stress, calcium disturbance, and acute membrane injury, whereas later sampling can evaluate inflammatory resolution, fibrosis, and remodeling. A single terminal time point may overlook transient oxidative changes or fail to distinguish delayed protection from simple postponement of injury.[10]

Translational Significance and Current Evidence Gaps

The isoprenaline model is useful for testing antioxidant mechanisms because oxidative stress is prominent and injury is reproducible. Nevertheless, positive results should be interpreted as protection against catecholamine-induced myocardial damage, not definitive treatment of human atherothrombotic infarction. Human infarction additionally involves plaque disruption, thrombosis, regional ischemia, reperfusion, comorbidities, and concurrent evidence-based therapy.[1]

Rats also synthesize vitamin C, and experimental doses of both antioxidants may be considerably higher than clinically achievable oral exposure. Differences in metabolism, treatment timing, and route of administration further restrict direct extrapolation. Pretreatment before isoprenaline administration primarily tests prevention and does not reproduce the clinical situation in which therapy usually begins after symptom onset.[19]

Direct evidence for α-lipoic acid in the isoprenaline model is less developed than the broader evidence for its antioxidant and mitochondrial effects. Similarly, evidence supporting vitamin C in this model is encouraging but remains largely preclinical. Carefully designed dose-response studies are needed to determine whether combined treatment improves cardiac structure and function beyond the effects of each compound separately.[8]

Future work should standardize isoprenaline dose, antioxidant formulation, administration schedule, sex and age of animals, tissue-collection time, and outcome definitions. Reporting should include randomization, blinded outcome assessment, sample-size justification, exclusions, and all prespecified outcomes. Replication across laboratories and comparison with coronary occlusion-reperfusion models would improve confidence in the biological relevance of the findings.[1]

Conclusion

Isoprenaline induces myocardial injury through a coordinated network of β-adrenergic overstimulation, excessive cardiac oxygen demand, catecholamine oxidation, calcium overload, mitochondrial failure, inflammation, and cardiomyocyte death. Oxidative stress is central to this process but cannot be separated from ionic, metabolic, and inflammatory disturbances.

α-Lipoic acid may provide protection by supporting mitochondrial metabolism, strengthening glutathione-dependent defenses, regulating redox-sensitive signaling, and limiting inflammatory and apoptotic pathways. Vitamin C may complement these actions through aqueous radical neutralization, antioxidant recycling, membrane protection, and preservation of endothelial redox function. Their combined use is physiologically credible, but genuine synergy cannot be assumed without direct comparison of individual and combined treatments using formal interaction analysis. Well-controlled experimental studies integrating biochemical, molecular, histological, and functional outcomes are required before the combination can be considered an established cardioprotective strategy.

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