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REVIEW

Translational science

Cation dyshomeostasis and cardiomyocyte necrosis: the Fleckenstein hypothesis revisited

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An ongoing loss of cardiomyocytes to apoptotic and necrotic cell death pathways contributes to the progressive nature of heart failure. The pathophysiological origins of necrotic cell loss relate to the neurohormonal activation that accompanies acute and chronic stressor states and which includes effector hormones of the adrenergic nervous system. Fifty years ago, Albrecht Fleckenstein and coworkers hypothesized the hyperadrenergic state, which accompanies such stressors, causes cardiomyocyte necrosis based on catecholamine-initiated excessive intracellular Ca^{2+} accumulation (EICA), and mitochondrial Ca^{2+} overloading in particular, in which the ensuing dysfunction and structural degeneration of these organelles leads to necrosis. In recent years, two downstream factors have been identified which, together with EICA, constitute a *signal–transducer–effector pathway*: (i) mitochondria-based induction of oxidative stress, in which the rate of reactive oxygen metabolite generation exceeds their rate of detoxification by endogenous antioxidant defences; and (ii) the opening of the mitochondrial inner membrane permeability transition pore (mPTP) followed by organellar swelling and degeneration. The pathogenesis of stress-related cardiomyopathy syndromes is likely related to this pathway. Other factors which can account for cytotoxicity in stressor states include: hypokalaemia; ionized hypocalcaemia and hypomagnesaemia with resultant elevations in parathyroid hormone serving as a potent mediator of EICA; and hypozincaemia with hyposelenaemia, which compromise antioxidant defences. Herein, we revisit the Fleckenstein hypothesis of EICA in leading to cardiomyocyte necrosis and the central role played by mitochondria.

Keywords Potassium • Magnesium • Calcium • Zinc • Selenium • Acute stressor states • Congestive heart failure • Neurohormonal activation

Introduction

An ongoing loss of cardiomyocytes via apoptotic and necrotic cell death pathways contributes to the progressive nature of heart failure. As depicted in *Figure 1*, apoptotic cells are rapidly scavenged by macrophages; they neither disintegrate nor lose their contents to stimulate the immune system. As a result, serum troponin levels are not elevated and a wound healing response is not invoked.[1–3] Dying necrotic cells, on the other hand, release troponins and other intracellular contents, which serve as danger signals to the immune system and chemoattractants that promote invasion of inflammatory cells to the site of injury. These cells, together with myofibroblasts, account for subsequent tissue repair. Foci of microscopic scarring are the final outcome. Hence, elevations in serum troponins and cardiac fibrosis are each footprints of cardiomyocyte necrosis. Scattered foci of fibrosis are found throughout both ventricles of the explanted failing human heart and are considered the major component of the pathological structural remodelling of myocardium.[4] This would not only implicate the importance of cardiomyocyte necrosis, but would also suggest it to be an ongoing process. The loss of cardiomyocytes and their replacement with stiff fibrillar collagen each contribute to the progressive failure of this muscular pump. Elevations in serum troponins are found in patients hospitalized because of their congestive heart failure (CHF) and are associated with an increased risk of morbidity and mortality from cardiovascular events.[5–14] In ambulatory asymptomatic elderly men, followed for 11 years in a community in Sweden, the appearance of elevated serum troponin predicted an increased risk of heart failure.[15] Factors other than overt ischaemia with a segment of infarcted

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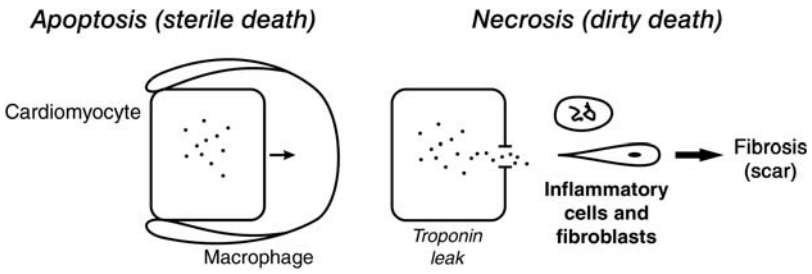

Figure 1 Heart failure involves an ongoing loss of cardiomyocytes to apoptosis and necrosis. See text.

myocardium can account for cardiomyocyte necrosis (vide infra). An understanding of pathophysiological mechanisms involved becomes essential to the optimal evaluation and management of these patients. Towards this end, the origins of the CHF syndrome provide crucial insights.

Congestive heart failure has its origins rooted in inappropriate neurohormonal activation. This includes the hypothalamic–pituitary–adrenal axis (HPA), the adrenergic nervous (ANS), and renin–angiotensin–aldosterone (RAAS) systems. Their effector hormones are cytotoxic to cardiomyocytes.[16–18] Some 50 years ago, Albrecht Fleckenstein and coworkers at the University of Freiburg im Breisgau hypothesized that hyperadrenergic state which accompanies stressor states, such as CHF, would lead to catecholamine-mediated excessive intracellular Ca^{2+} accumulation (EICA), particularly involving cardiac mitochondria. The ensuing dysfunction of Ca^{2+} overloaded mitochondria, coupled with the diminished synthesis of high-energy phosphate and structural degeneration of these organelles, would lead to cardiomyocyte necrosis. They validated their hypothesis using isoproterenol-induced cardiac injury in rodents in which cotreatment with a calcium-channel blocker, verapamil, proved cardioprotective.[19,20] Later, others confirmed this paradigm and provided further insights into the adverse consequences of elevated plasma epinephrine levels (5000 pg/mL) comparable with those found in man during acute and chronic stressor states.[18,21–24] Today, the importance of catecholamine excess that accompanies marked emotional stress or acute stressor states, such as head trauma or subarachnoid haemorrhage, is now recognized as leading to stress-related cardiomyopathy syndromes (e.g. apical ballooning or Takotsubo cardiomyopathy).[25]

In recent years, two other factors, together with EICA, were identified to be major participants in a *signal–transducer–effector pathway* to cardiomyocyte necrosis during acute or chronic hyperadrenergic states (see *Figure 2*). This includes the genesis of oxidative stress, where the rate of reactive oxygen and nitrogen species generation overwhelms their rate of elimination by endogenous antioxidant defences, invoked in response to EICA. Second, the role of the mitochondrial inner membrane permeability transition pore (mPTP) opening which leads to organellar dysfunction, osmotic swelling, and ultimate structural degeneration of these organelles. Other pathophysiological responses that accompany catecholamine excess and which extend beyond the importance of Ca^{2+} overloading can also be cytotoxic. They cannot be overlooked and include a dyshomeostasis of essential cations which are manifested as hypokalaemia, ionized hypomagnesaemia and hypocalcaemia, hypozincaemia, and hyposelenaemia. Herein, we introduce and highlight this broader perspective of cation dyshomeostasis in revisiting the Fleckenstein hypothesis and cardiomyocyte necrosis.

Acute stressor states and cation dyshomeostasis

Neurohormonal activation

Acute stressor states are broadly referred to as representing acute bodily injury in one form or another. For example, they include: acute myocardial infarction; major cardiac or noncardiac surgery; thermal or electrical burns; head or musculoskeletal trauma; and subarachnoid haemorrhage or intracerebral bleed. An acute systemic inflammatory response invoked by sepsis or diabetic ketoacidosis is another example. Acute stressor states are inextricably linked to neurohormonal activation involving the HPA axis as well as the ANS and RAAS, and whose effector hormones are integral to acute stressor state-mediated homeostatic responses. Catecholamines, parathyroid hormone (PTH), angiotensin II, and endothelin-1 account for *homeostasis gone awry to beget dyshomeostasis* at cellular and molecular levels involving the heart and systemic organs. This includes a dyshomeostasis of mono- and divalent cations. At the time of or shortly after hospital admission, a dyshomeostasis of a whole host of electrolytes and trace elements are manifested contemporaneously in critically ill patients (*Figure 3*). These effector hormones orchestrate the concordant appearance of hypokalaemia, ionized hypocalcaemia and hypomagnesaemia, hypozincaemia and hyposelenaemia. The shift in electrolytes from blood to soft tissues accounts for ionized hypocalcaemia and hypomagnesaemia which will invoke secondary hyperparathyroidism (SHPT) with the parathyroid glands' elaboration of the calcitropic PTH (*Figure 4*) seeking to restore the homeostasis of these circulating divalent cations through bone mineral resorption. Intracellular cation shifts, particularly catecholamine- and PTH-mediated EICA, converge on mitochondria to induce oxidative stress and raise the opening potential of their inner membrane mPTP (*Figure 2*). The ensuing loss of intracellular cationic homeostasis and diminished ATP synthesis, together with osmotic swelling of mitochondria, lead to organellar degeneration.

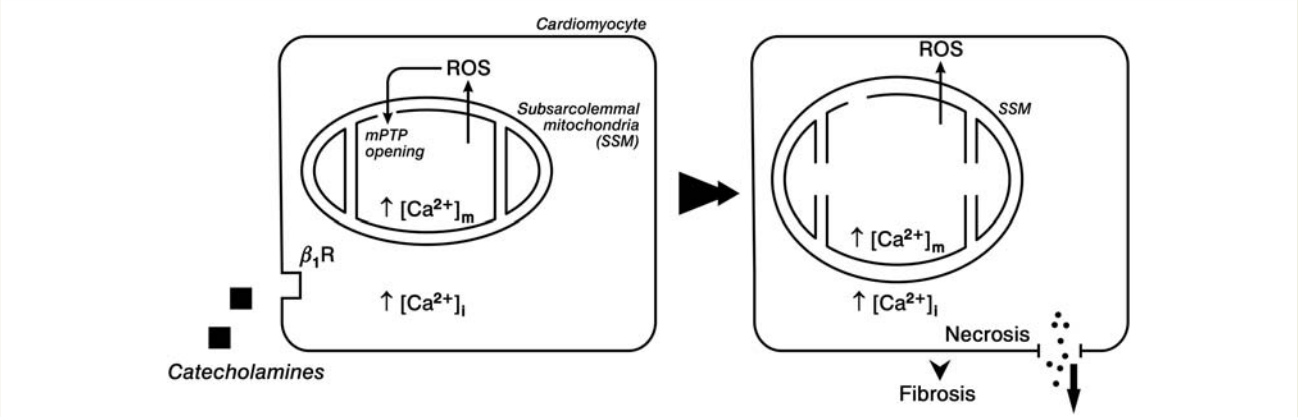

Figure 2 Catecholamine-mediated cellular and subcellular Ca^{2+} overloading with induction of oxidative stress and reactive oxygen species generation and opening of the mitochondrial inner membrane permeability transition pore that leads to solute entry, osmotic swelling and structural degeneration of these organelles. Cell death follows with a leak of intracellular troponins, which raise serum troponin levels, and ultimate appearance of replacement fibrosis, or scarring.

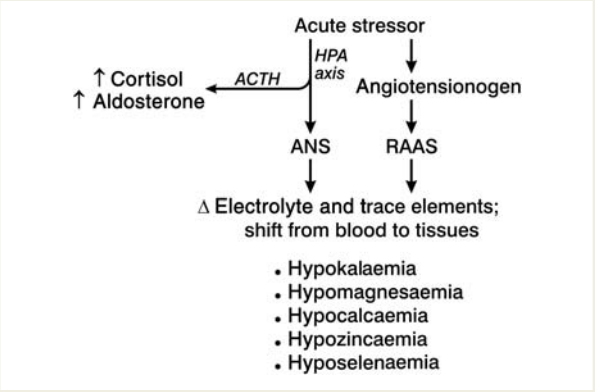

Figure 3 An acute stressor state, such as bodily injury, activates the hypothalamic–pituitary–adrenal axis with resultant release of adrenocorticotropin hormone, which promotes the adrenals' release of cortisol and aldosterone, and catecholamines from the adrenal medulla. The acute phase reactant, angiotensinogen, is released by the liver during stressor states and is accompanied by activation of the renin–angiotensin–aldosterone system . In turn, elevated plasma catecholamines, norepinephrine, and epinephrine, promote a coordinated cation translocation from the vascular space to tissue compartment accounting for a concordant fall in their serum concentrations and presenting as hypokalaemia, ionized hypocalcaemia and hypomagnesaemia, hypozincaemia and hyposelenaemia.

Cardiomyocyte necrosis follows with the leakage of troponins ultimately appearing in the circulation as biomarkers confirmatory of necrosis.

Hypokalaemia

Catecholamines promote hypokalaemia. Struthers *et al.*[26–28] administered intravenous epinephrine to normal human volunteers and demonstrated a prompt and marked fall in serum K^+ of 0.8 ± 0.19 mEq/L (from 4.0 to 3.2 mEq/L) which was prevented by a β_2-adrenergic receptor blocker. A simultaneous fall in serum Mg^{2+} and Ca^{2+} also occurred. In patients with acute bodily injury accompanied by haemorrhagic shock, endogenous plasma catecholamines are markedly elevated to promote arteriolar vasoconstriction and in so doing raise fallen arterial pressure. When these levels are further elevated by pharmacological doses of exogenous norepinephrine, epinephrine, or dopamine, given to further raise blood pressure from shock levels, the reductions in serum K^+ (<3.0 mEq/dL) and Mg^{2+} (<1.5 mg/dL) can be more profound and lead to serious atrial and malignant ventricular arrhythmias.[29]

The underlying K^+ balance prior to bodily injury determines the severity of the ensuing hypokalaemia during an acute stressor state. Pretreatment of normal volunteers with a thiazide diuretic predisposed them to marked hypokalaemia in response to epinephrine infusion.[30] Spironolactone (Spiro), an aldosterone antagonist, was protective against hypokalaemia in this setting.[31] Patients with arterial hypertension or CHF who are receiving long-term thiazide or loop diuretic treatment, respectively, may have marginal K^+ and Mg^{2+} reservoirs, which are then further compromised by a hyperadrenergic state that accompanies bodily injury or acute myocardial infarction leading quickly to marked hypokalaemia and hypomagnesaemia with consequent QTc prolongation and a greater propensity for arrhythmias. Inhaled albuterol can likewise predispose to hypokalaemia and hypomagnesaemia in normal volunteers and those receiving diuretics.[27] Chronic excessive use of β_2 receptor agonists also lead to marked hypokalaemia and arrhythmias and injury to the heart and skeletal muscle.[32] Drug-induced prolongation of myocardial repolarization, as reflected in the lengthening of the QTc interval of the electrocardiogram, usually accompany certain antibiotics, antidepressants, and antipsychotics.[33,34] Prolongation of the QTc interval enhances the risk of polymorphic ventricular tachycardia, also known as *torsades de pointes*. Risk factors for drug-related QTc prolongation include hypokalaemia, sympathomimetics, and the concomitant

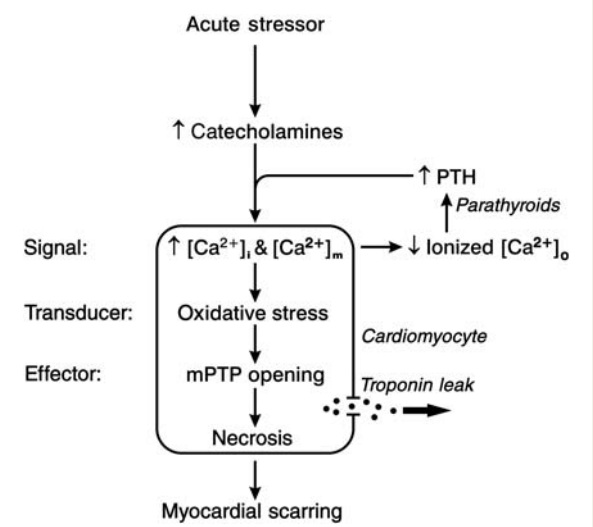

Figure 4 An acute stressor state with elevated circulating catecholamines is responsible for intracellular Ca^{2+} overloading with a subsequent fall in plasma ionized $[Ca^{2+}]_o$, which in turn provokes the parathyroid glands to release parathyroid hormone, a calcitropic hormone, also contributing to intracellular Ca^{2+} overloading. In cardiomyocytes this is accompanied by the induction of oxidative stress, which leads to the opening of the mitochondrial permeability transition pore and osmotic injury of these organelles. The necrosis of cardiomyocytes follows accompanied by the leak of intracellular troponins into the interstitial space accounting for the ultimate rise in plasma troponins. Cardiac myocytes lost to necrosis are replaced by fibrous tissue, or scarring, which preserves the structural integrity of the myocardium. Adapted from Whitted AD *et al. Am J Med Sci.* 2010;340:48–53.

administration of several of these agents.[35] Furthermore, hypokalaemia has been associated with cardiomyocyte necrosis and resultant cardiac pathology.[36]

Hypomagnesaemia

Dietary Mg^{2+} deficiency can cause cardiovascular lesions that eventuate in heart failure.[37,38] Elevations in plasma catecholamines associated with an acute stressor state are accompanied by hypomagnesaemia which is related to a cyclic AMP-mediated rise in intracellular Mg^{2+}, together with increased lipolysis and Mg^{2+} binding to free fatty acids.[39] Hypomagnesaemia is common in critically ill children and adults with such predisposing risk factors as hypokalaemia, hypocalcaemia, thiazide and loop diuretics, and sepsis. The hypomagnesaemia prevalent on admission in critically ill patients may worsen during prolonged hospital stay due to ongoing excretory losses and reduced Mg^{2+} intake.[40–42] Moreover, atrial and ventricular arrhythmias appear when hypomagnesaemia is of moderate to marked severity (<1.70 mg/dL).[43–45]

Concurrent hypokalaemia and hypomagnesaemia

Contemporaneous hypokalaemia and hypomagnesaemia are common in critically ill patients. The interactions of K^+ and

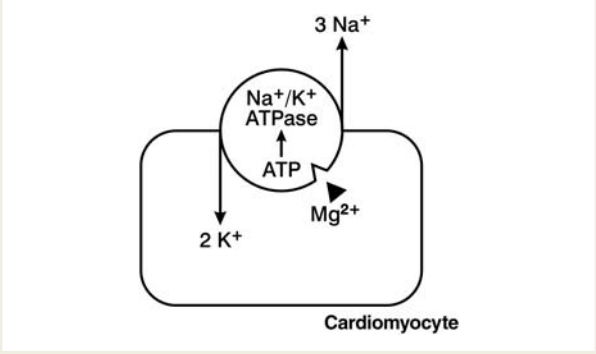

Figure 5 The sodium pump of the cardiomyocyte is an energy consuming, Mg^{2+}-dependent Na^+/K^+ ATPase which is responsible for the extrusion of three Na^+ ions and entry of two K^+ ions. Pump activity falters with Mg^{2+} deficiency accompanied by reduced intracellular K^+ and prolongation of the QTc interval of the electrocardiogram. In the presence of hypokalaemia and hypomagnesaemia, digoxin, a Na^+/K^+ ATPase inhibitor, would further reduce intracellular K^+ to raise the potential for arrhythmias.

Mg^{2+} are multifactorial and complex, including the importance of Mg^{2+} deficiency that interferes with K^+ retention.[46] The ability to successfully correct hypokalaemia mandates the simultaneous reversal of hypomagnesaemia.[47–49] The cell membrane's Na^+/K^+-ATPase pump maintains the crucial electrochemical K^+ gradient between high-intracellular K^+ concentration with lower K^+ concentration of the extracellular compartment (see *Figure 5*). Activated by Mg^{2+} this pump requires ATP as its energy source and hence Mg^{2+} participates in maintaining intracellular K^+, which falters during Mg^{2+} deficiency with suboptimal amounts of K^+ pumped into cells. As a result, Mg^{2+} deficiency contemporaneously begets K^+ deficiency. Digoxin, a Na^+/K^+ ATPase inhibitor, can worsen this dyshomeostasis by limiting renal tubular reabsorption of Mg^{2+} and thereby raising urinary Mg^{2+} excretion which exacerbates hypomagnesaemia and further predisposes to arrhythmias in this setting.[50] In order to resolve hypokalaemia, the Mg^{2+} deficiency must first or simultaneously be restored. In the absence of gastrointestinal losses or diuretic and digoxin usage, hypomagnesaemia and hypokalaemia due to impaired renal tubular reabsorption, in the form of urinary K^+ and Mg^{2+} wasting, must be considered. Inheritable renal tubular disorders, such as the Gitelman syndrome in adults and Bartter syndrome in children, should be addressed when prompt resolution of these cations using oral Mg^{2+} and K^+ supplements proves difficult to achieve.[51] Regular serum electrolyte measurements should be augmented with serial ECG monitoring of the QTc interval, a useful biomarker of intracellular K^+ and Mg^{2+} levels. QTc prolongation (>460 ms) demonstrates their deficiency while its normalization serves to address the adequacy of their cellular replacement. The attainment of QTc of <460 ms with these supplements may require several additional days compared with the relatively rapid return of their normal serum levels.

Hypocalcaemia and intracellular Ca^{2+} overloading

Reductions in plasma ionized $[Ca^{2+}]_o$ are commonly found in the emergency department and intensive care units in patients having an acute stressor state with elevated plasma catecholamines (*Figure 4*). The fall in $[Ca^{2+}]_o$ correlates with the severity of the hyperadrenergic state and, in turn, the severity of illness. Ionized hypocalcaemia serves as an in-hospital predictor of survival.[52–61] Hypoalbuminaemia can contribute to reduced total Ca^{2+} concentration. In response to hypocalcaemia, the Ca^{2+}-sensing receptor of the parathyroid glands provokes stimulated secretion of PTH. The ensuing SHPT seeks to restore extracellular Ca^{2+} homeostasis by promoting the resorption of bone Ca^{2+} and increased Ca^{2+} absorption from the gut and kidneys. When hypocalcaemia is associated with hypomagnesaemia, PTH secretion may be impaired but can be rapidly resolved by reversing hypomagnesaemia.

The appearance of acute ionized hypocalcaemia in critically ill patients is caused by a shift in Ca^{2+} from the circulating pool to the intracellular compartment of various tissues, including the heart and skeletal muscle. This cation shift occurs in response to catecholamine-induced intracellular Ca^{2+} overloading followed by PTH-mediated excessive Ca^{2+} entry (*Figure 4*). Thus, catecholamine- and PTH-facilitated intracellular Ca^{2+} overloading of cardiomyocytes, in keeping with the Fleckenstein hypothesis, converge into mitochondrial Ca^{2+} overloading and is coupled to the induction of oxidative stress. The ensuing necrotic death of cardiomyocytes is followed by tissue repair and a consequent replacement fibrosis. Such scarring preserves the structural integrity of the myocardium. However, this structural remodelling has adverse consequences. These include compromised myocardial stiffness and ventricular function which collectively serve as substrate for reentrant arrhythmia.

The catecholamine-induced disintegration of necrotic cardiomyocytes is accompanied by the release of troponins, an intracellular enzyme that plays a crucial role in revealing myocardial injury (*Figure 2*). Catecholamine-induced cardiomyocyte necrosis with increased plasma troponin levels occur in critically ill patients, including those having sepsis, haemorrhagic shock, subarachnoid haemorrhage, trauma, gastrointestinal bleeding, or pulmonary embolus.[62–65] The levels to which plasma troponins rise in such patients, however, do not reach the more marked elevations seen with the segmental loss of infarcted myocardium that accompanies an acute reductions in coronary blood flow due to a thrombosed coronary artery.

Hypozincaemia

Hypozincaemia appears in critically ill patients, including those having an acute myocardial infarction[66–73] where it persists during much of the first week and then slowly recovers.[74,75] It also appears during week 1 following major trauma and is related to excessive urinary excretion and fluid losses, reduced Zn^{2+} intake and preferential redistribution of Zn^{2+} to injured tissues.[76] Tissue Zn^{2+} contributes to antioxidant defences, and are integral to wound healing.[77–79] Hypozincaemia is frequently associated with hyposelenaemia.[77,80,81]

Hyposelenaemia

Hyposelenaemia has been identified on admission in patients with an acute myocardial infarction, where it correlates with the rise in serum troponin levels.[82] In critically ill patients having the systemic inflammatory response syndrome, hyposelenaemia is accompanied by reduced plasma Se-glutathione peroxidase (GSHPx) activity.[83] Since thyroid hormone is a selenoprotein, thyroid function can be compromised with hyposelenaemia.

Summary

The complex dyshomeostasis of electrolytes and trace elements that occurs with acute stressor states has broad and diverse pathophysiological sequelae, including cardiomyocyte necrosis. To minimize adverse cardiovascular consequences during hyperadrenergic states, systematic and serial surveillance of serum K^+, Mg^{2+}, and Ca^{2+} is warranted. Complementary protective measures should include QTc interval monitoring with serial ECG, a biomarker of myocardial repolarization. Prolonged QTc, due to reduced intracellular K^+ and Mg^{2+} or to drug therapy, raises the vulnerability of the heart to atrial and/or ventricular arrhythmias. The maintenance of serum K^+ and Mg^{2+} within the strictly defined narrow physiological threshold (i.e. $K^+ \geq 4.0$ mEq/L and $Mg^{2+} \geq 2.0$ mg/dL) will inevitably prove most effective in preventing arrhythmias. An awareness of hypozincaemia and hyposelenaemia also broadens our clinical perspective on the acute stressor state paradigm to include their deleterious impacts on the compromised efficiency of metalloenzyme-based antioxidant defences to combat oxidative stress.

Chronic stressor states and cation dyshomeostasis

Chronic stressor states include: a failure of the heart, kidneys, lungs, or liver, irrespective of aetiological origins; and chronic inflammatory diseases, such as rheumatoid arthritis, psoriasis, and inflammatory bowel disease. We now focus on the chronic neurohormonal activation involving the HPA axis, ANS, and RAAS which are integral pathophysiological features of CHF, and which occurs irrespective of its aetiological origins or patient age. Elevated plasma levels of cortisol, renin activity, angiotensin II, aldosterone, epinephrine, norepinephrine, and endothelin–1 are each found in CHF.[84–88]

Hypokalaemia and hypomagnesaemia

Renin–angiotensin–aldosterone system activation in patients with systolic or diastolic heart failure leads to a salt-avid state with Na^+ and water retention that eventuates in the appearance of symptoms and signs of the CHF syndrome. Urinary and faecal excretion of K^+ and Mg^{2+} are increased during CHF based on the endocrine-mediated actions of circulating aldosterone acting at these sites, where high-density aldosterone receptor binding occurs. The loss of these cations is accentuated by loop diuretics commonly used in the management of CHF.[47,89] Chronic hypomagnesaemia is frequently associated with hypokalaemia and hypocalcaemia and portends an adverse prognosis.[90] Loop as well as thiazide diuretics promote excessive urinary loss of K^+ and Mg^{2+} that

can lead to both hypokalaemia and hypomagnesaemia. Combining either of these diuretics with Spiro preserves K^+ and Mg^{2+} homeostasis,[30] provided renal function is not markedly impaired (serum creatinine <2.0 mg/dL) and K^+ supplements are discontinued.

The importance of hypokalaemia on patient mortality has been well documented. The Digitalis Investigative Group (DIG) trial database involving more than 7700 patients revealed that in ambulatory patients having either systolic or diastolic heart failure, serum K^+ <4.0 mEq/L and Mg^{2+} <2.0 mg/dL were associated with increased mortality.[91,92] The same was true in patients with heart failure having associated chronic kidney disease.[93] This database also revealed the adverse impact of loop diuretics on death, cardiovascular mortality, and heart failure-related hospitalization in ambulatory patients, including the elderly.[94,95] This raises the prospect that prolonged routine use of a potent loop diuretic, in the absence of symptoms and signs of salt avidity, can be quite deleterious and should be discontinued and milder diuretics implemented, if necessary, in salt-sensitive patients.[96] However, the loop diuretic can be reinstituted, if and when the patient is again avidly and persistently retaining Na^+ and water.

In the Study of Left Ventricular Dysfunction (SOLVD) trial with a cohort of more than 6700 patients, such adverse events were not seen with potassium-sparing diuretics, such as Spiro, amiloride, or triamterene. Indeed, these agents may be associated with reduced risk of all-cause mortality or death from or hospitalization for progressive heart failure.[97–99] Spiro, an aldosterone receptor antagonist, conserves both K^+ and Mg^{2+}. In the Randomized Aldactone Evaluation (RALES) trial the efficacy and safety of Spiro, when combined with an ACE-Inhibitor or angiotensin receptor blocker and a loop diuretic, was demonstrated and included a 30% risk reduction for all-cause and cardiovascular-related mortality and sudden cardiac death and cardiovascular morbidities.[99]

Ionized hypocalcaemia and intracellular Ca^{2+} overloading

The secondary aldosteronism of CHF in man leads to increased faecal and urinary Ca^{2+} excretion and consequent ionized hypocalcaemia and, in turn, SHPT with elevated plasma PTH levels.[80,100–103] As noted earlier, dyshomeostasis of divalent cations frequently occurs in patients hospitalized with decompensated biventricular failure having a dilated cardiomyopathy. Elevated plasma PTH levels and SHPT are also found in patients with pulmonary hypertension or obstructive airway disease,[104,105] in which RAAS activation with secondary aldosteronism is expected due to reduced systemic blood flow that includes renal perfusion. This hormonal profile is found in patients with primary aldosteronism,[106–109] where aberrations in serum ionized and total Ca^{2+}, together with elevated PTH, are normalized by either Spiro or adrenal surgery.[108,109] Furthermore, elevated PTH is a known stimulus to adrenal aldosterone production and can further account for elevated plasma aldosterone levels. In patients with primary hyperparathyroidism, preoperative PTH levels in excess of 100 ng/mL are independent predictors of abnormally elevated plasma aldosterone levels.[110] The impact of chronic aldosteronism on the increased incidence of adverse cardiovascular outcomes in patients with primary hyperparathyroidism remains uncertain.[111] However, experimental findings congruently point towards the importance of PTH-mediated intracellular Ca^{2+} overloading and induction of oxidative stress as major pathogenic events accounting for adverse myocardial remodelling, as contrasted to elevations in circulating aldosterone, *per se*.[112–114]

Abnormal elevations in serum PTH (>65 pg/mL), a calcitropic hormone and mediator of EICA in cardiomyocytes and mitochondria,[112,115,116] are found in patients hospitalized with decompensated heart failure and those awaiting cardiac transplantation.[100,103,117,118] In outpatients having heart failure, elevated serum PTH levels are also identified and serve as an independent predictor of CHF and the need for hospitalization.[119–121] Plasma PTH levels were shown to be an independent risk factor for mortality and cardiovascular events in patients undergoing coronary angiography in Austria,[122] and increased risk for cardiovascular mortality and the risk of heart failure were predicted in a community-based cohort of elderly men followed longitudinally for 8 years or more in Sweden.[123,124] We found SHPT to be especially prevalent in African-Americans (AA) with protracted decompensated biventricular failure, where chronic elevations in plasma aldosterone account for symptoms and signs of CHF.[103] Secondary hyperparathyroidism is also related to the prevalence of hypovitaminosis D in AA with CHF.[103] The increased melanin content of darker skin in AA serves as a natural sunscreen. Accordingly, the prevalence of hypovitaminosis D, often of marked severity (<10 ng/mL), compromises Ca^{2+} homeostasis predisposing AA to hypocalcaemia and consequent SHPT.[103,125,126] Vitamin D deficiency is also common in Caucasians and Asians with heart failure.[119,127–129]

Other factors which may be associated with compromised Ca^{2+} stores and contribute to the appearance of SHPT, especially in AA with CHF, include: reduced dietary Ca^{2+} intake because of lactose intolerance and an active avoidance of dairy products rich in Ca^{2+}[130]; and a preference for a high-Na^+ diet that enhances urinary Ca^{2+} excretion. A high-salt diet and consequential hypercalciuria is well known for predisposing patients to ionized hypocalcaemia and SHPT with resorption of bone which is invoked to restore extracellular Ca^{2+} homeostasis. Over time, osteopenia and osteoporosis appear as an adverse outcome to SHPT invoked by the hypercalciuria of long-term dietary Na^+ excess further predisposing to atraumatic bone fractures.[131,132] Patients with heart failure have reduced bone density, which is related to SHPT and vitamin D deficiency coupled with reduced physical activity that may be a cofactor of their effort intolerance due to symptomatic failure.[100,117,133–137] The risk of such fractures is increased in elderly patients with heart failure,[138] where SHPT may be contributory, and which appears to be preventable when Spiro is combined with today's standard of care.[139]

Elevations in serum troponins, biomarkers of cardiomyocyte necrosis, but not due to acute MI or renal failure, are found in patients hospitalized because of their decompensated heart failure and are associated with increased in-hospital and overall cardiac mortality.[5–14] The role of intracellular Ca^{2+} overloading and oxidative stress, induced by neurohormonal activation that includes calcitropic hormones, catecholamines

and PTH, in promoting myocardial cell loss in these patients is not absolutely clear, but must be explored. An ongoing loss of cardiomyocytes contributes to the progressive nature of heart failure.

Zn^{2+} and Se^{2+} dyshomeostasis

In addition to hypokalaemia, ionized hypocalcaemia and hypomagnesaemia that accompany increased urinary and faecal losses of these divalent cations with the aldosteronism of CHF, there also is a concomitant dyshomeostasis of Zn^{2+} with hypozincaemia.[81,140] Furthermore, urinary Zn^{2+} excretion is increased in response to angiotensin-converting enzyme inhibitor or angiotensin receptor antagonist, commonly prescribed agents in the management of patients with CHF and where hypozincaemia is associated with abnormalities in taste (or dysgeusia).[141,142] Serum Zn^{2+} and Se^{2+} levels are reduced in AA patients.[80,102] This includes those with decompensated failure and compensated failure, as well as with heart disease but without heart failure. Interactions between Zn^{2+} and Se^{2+} have been reported.[143] Underlying causes for the simultaneous deficiencies of these divalent cations in AA, including inadequate dietary intake, are presently uncertain.

The prooxidant effect representing intracellular Ca^{2+} overloading that accompanies elevations in either plasma catecholamines or PTH is intrinsically coupled to Zn^{2+} entry acting as an antioxidant.[115,116,144,145] Although less robust, Zn^{2+} entry is known to occur via L-type Ca^{2+} channels whereas more substantive amounts ingress by Zn^{2+} transporters activated by oxidative stress. The release of inactive Zn^{2+} bound to metallothionein-1 contributes to increased cytosolic-free levels of Zn^{2+}, which can also be achieved by a $ZnSO_4$ supplement or Zn^{2+} ionophore.[145,146] These cumulative salutary observations raise the therapeutic prospect that cation-containing *nutriceuticals* capable of favourably influencing extra- and intracellular Ca^{2+} and Zn^{2+} equilibrium, which is pivotal to combating oxidative injury and promoting repair, could attenuate or even prevent cardiomyocyte necrosis and myocardial scarring.

Selenium is a cofactor of antioxidant selenoenzymes, such as GSH-Px and thioredoxin reductase, that promote optimal antioxidant/oxidant balance.[147] Monitoring serum Se levels, Se-dependent enzymatic activities, and Se-GSH-Px mRNA expression are clinically useful in addressing optimal Se supplementation.[148,149] Appearance of a dilated cardiomyopathy in greater abundance has been reported in general populations, in which dietary Se^{2+} deficiencies are found, such as in the Se-poor soil of the Keysan Province of China, or when parenteral nutrition was inadvertently deficient in Zn and/or Se.[150–152] The selenium-deficiency-induced cardiomyopathy is often reversible with Se^{2+} replacement.[153]

Summary

Thus, neurohormonal activation that accompanies CHF is comparable with acute stressor states (*Table 1*). Together with the adverse impact of loop diuretics, there is a concerted and contemporaneous complex dyshomeostasis of K^+, Mg^{2+}, and Ca^{2+} associated with adverse pathophysiological consequences. Compromised Ca^{2+} stores related to excretory losses and/or altered dietary intake, together with vitamin D deficiency, predispose to SHPT with compromised cardiomyocyte survival and impaired skeletal health.

Table 1 A common signal–transducer–effector pathway to cardiomyocyte necrosis in acute and chronic stressor states

	Stressor state	
	Acute	Chronic
Neurohormonal activation		
HPA axis	+	+
ANS	+	+
RAAS	+	+
Cation dyshomeostasis		
↑ $[Ca^{2+}]_i$ & $[Ca^{2+}]_m$	+	+
↑ $[Zn^{2+}]_i$ & $[Zn^{2+}]_m$	±	+
Oxidative stress > antioxidant defences	+	+
mPTP opening	+	+

Taken together, the multitude of evidence gathered to date congruently supports the Fleckenstein hypothesis which invokes catecholamine- and PTH-mediated intracellular Ca^{2+} overloading as the most tenable mechanism leading to the induction of oxidative stress, where ROS and RNS, primarily derived from mitochondria in cardiomyocytes and membrane-bound NADPH oxidase in vascular tissue, overwhelm cellular antioxidant defences. This scenario anticipates the question whether ensuing adverse consequences are the result of an excessive generation of prooxidants or due to compromised endogenous antioxidant defences, or both. Zn^{2+} supplementation, serving as antioxidant, has shown promise in enhancing antioxidant defences in experimental animals receiving aldosterone/salt treatment or having streptozocin-induced diabetes.[115,145,146,154] A polynutrient supplement, however, which includes these cations and vitamin D, at a minimum, will likely be necessary. Promising results with a polynutrient supplement have been reported in critically ill patients, including those with heart failure.[155–159]

Summary and conclusions

Acute and chronic stressor states are each accompanied by neurohormonal activation that includes the ANS. As Fleckenstein and coworkers originally envisaged, the hyperadrenergic state is accompanied by cardiomyocyte Ca^{2+} overloading, particularly involving their mitochondria, with resultant dysfunction and disintegration of the organelles and ensuing necrotic cell death. More recent studies have identified subsarcolemmal mitochondria-based induction of oxidative stress and opening of their inner membrane mPTP as other major components of the pathophysiological signal–transducer–effector pathway to cardiomyocyte necrosis which eventuates in the release of troponins causing elevated serum troponins and a consequent wound healing response leading to scattered foci of microscopic scarring. Fibrosis is a major component to the adverse structural remodelling of failing

myocardium and whose ongoing appearance accounts for the progressive failure of this normally efficient muscular pump.

Furthermore, neurohormonal activation, including HPA axis, ANS and RAAS, and their effector hormones, orchestrate the concordant appearance of hypokalaemia, ionized hypocalcaemia and hypomagnesaemia, hypozincaemia and hyposelenaemia, and is based on the coordinated translocation of cations to injured tissues. Intracellular cation shifts adaptively regulate the equilibrium between prooxidants and antioxidants, a critical determinant of cardiomyocyte survival. The intrinsically coupled dyshomeostasis of Ca^{2+} and Zn^{2+}, representing prooxidant and antioxidant, respectively, can be uncoupled in favour of increased intracellular-free Zn^{2+} and antioxidant defences. In so doing, cardiomyocytes that are on the brink of necrotic death can be rescued. The use of nutriceuticals to achieve these lofty goals ought to be considered as complementary to today's standard of care using pharmaceuticals alone.

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