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RESEARCH ARTICLE

Effect of dietary selenium on the progression of heart failure in the ageing spontaneously hypertensive rat

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Oxidative stress has been directly implicated in hypertension and myocardial remodelling, two pathologies fundamental to the development of chronic heart failure. Selenium (Se) can act directly and indirectly as an antioxidant and a lowered Se status leads to a higher risk of cardiovascular disease. This study examined the role of Se on the development of hypertension and subsequent progression to chronic heart failure in spontaneously hypertensive rats (SHR). Three dietary groups were studied: (i) Se-free; (ii) normal Se (50 μg Se/kg food); and (iii) high Se (1000 μg Se/kg food). Systolic blood pressure and echocardiography were used to detect cardiac changes *in vivo*. At study end, cardiac tissues were assayed for glutathione peroxidase activity, thioredoxin reductase activity, and protein carbonyls. The major finding of this study was the high heart failure-related mortality rate in SHRs fed an Se-free diet (70%). Normal and high levels of dietary Se resulted in higher survival rates of 78 and 100%, respectively. Furthermore, high dietary Se was clearly associated with lower levels of cardiac oxidative damage and increased antioxidant expression, as well as a reduction in disease severity and mortality in the SHR.

Keywords:
Glutathione peroxidase / Heart failure / Hypertension / Selenium / Thioredoxin reductase

1 Introduction

Increasing evidence supports a role for oxidative stress in the pathogenesis of hypertension and the subsequent progression to compensated hypertrophy and end-stage heart failure [1–6]. Oxidative stress can be countered by increasing the exogenous supply of antioxidants or increasing the cellular expression of endogenous antioxidants. The role of selenium (Se) as an antioxidant has been well established and a lowered Se status has been associated with an increased risk of cardiovascular disease, including chronic heart failure (CHF) [3, 4]. An important biological function of Se is mediated through the antioxidant selenoproteins glutathione peroxidase (GPx) and thioredoxin reductase (Thx-Red). The cardiovascular benefits of Se have been consistently demonstrated in animal studies, with dietary Se supplementation prior to ischemia-reperfusion injury shown to result in improved cardiac functional recovery [7–10], reduced incidence of reperfusion arrhythmias [10], and preservation of ventricular ultrastructure [11]. As expected, these studies reported increases in selenoprotein expression and activity and a concomitant reduction in markers of oxidative injury. While Se supplementation has shown benefit in short-term studies, less is known on the longer term affects in the chronic progression to heart failure.

Control of dietary Se effectively modulates both the expression and activity of the selenoproteins, such as GPx and Thx-Red, and this has been consistently demonstrated by our laboratory and others [9, 11–14]. The aim of the

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Abbreviations: CHF, chronic heart failure; **DTNB,** Dithiobis (2-nitrobenzoic acid); **FS,** fractional shortening; **GPx,** glutathione peroxidase; **LV,** left ventricular; **LVEF,** left ventricular ejection fraction; **ROS,** reactive oxygen species; **SBP,** systolic blood pressure; **Se,** selenium; **SHR,** spontaneously hypertensive rat; **Thx-Red,** thioredoxin reductase; **WKY,** Wistar-Kyoto

current study was to evaluate the significance of dietary Se on the development of hypertension and subsequent progression to heart failure, using the spontaneously hypertensive rat (SHR). The SHR strain was developed from outbred Wistar-Kyoto (WKY) rats and WKY rats are used as normotensive controls [15, 16]. The SHR strain develops hypertension at 7–15 wk of age, with systolic blood pressure (SBP) reaching a plateau at around 200 mmHg. Myocardial hypertrophy develops as a consequence of chronic pressure overload and the onset of heart failure usually occurs at 16–18 months of age [15]. SHRs exhibit several features of the human disease including systemic vascular congestion, ventricular dilation, and decreased left ventricular (LV) ejection fraction [16]. Clinical signs of heart failure include dyspnea, cyanosis, lethargy, weight loss, and peripheral oedema [17–19]. Our study also aimed to investigate the biochemical mechanisms underpinning the physiological effects, by measuring selenoprotein activity and oxidative injury in SHR rats fed a Se-depleted diet.

2 Materials and methods

2.1 Animals, experimental diet, and experimental design

Male SHR and WKY rats (12 wk of age) were obtained from the Animal Resources Centre, Western Australia. All experimental procedures used in this study were approved by the Griffith University Animal Ethics Committee, which acts in accordance with the *Australian Code of Practice for the Care and Use of Animals for Scientific Purposes* and the *NIH Guide for the Care and Use of Laboratory Animals* (NIH publication No. 85–23, revised 1996). The animals were fed standard rat pellets until they were 65 wk of age, then assigned to one of three dietary groups and fed an experimental diet for 7 months (until 95 wk of age).The basal diet was Se free, containing 30% torula yeast, 59% sucrose, 5% coconut oil (vitamin E free), 5% premixed minerals, and 1% premixed vitamins (MP Biomedicals, Seven Hills, Australia). A diet containing no Se was used to induce severe Se deficiency (Se-free: SHR: $n = 10$; WKY: $n = 6$). A diet containing 50 μg Se/kg food was used to reflect the most common intake in humans (normal Se: SHR: $n = 9$; WKY: $n = 6$), and a high Se diet of 1000 μg Se/kg food was used to reflect a high Se intake (high Se: SHR: $n = 9$; WKY: $n = 6$) [10]. Animals consumed approximately 10–20 g of food *per* day, which resulted in a daily intake of 1–2 μg Se/kg bodyweight for the 50 μg Se/kg group and 20–40 μg/kg bodyweight for the high Se group. The current recommended daily intake for humans is approximately 1 μg/kg of bodyweight [45].

Throughout the 7-month period, SBP measurements, echocardiography, and body weight measurements were performed once a month for SHRs and every 3 months for WKY rats. The study was terminated when the animals were 22 months of age when the animals were euthanased with an i.p. injection of sodium pentobarbitone (200 mg/kg).

2.2 SBP measurements

SBP was measured by the tail-cuff method, using a ML125 NIBP Controller with a tail pulse transducer (MLT1125R) and an inflatable pressure cuff, connected to a PowerLab data acquisition unit (ADInstruments, Sydney, Australia). The tail cuff was inflated to a maximum inflation pressure of 200 mmHg for WKY rats and 280 mmHg for SHRs and SBP was determined as the pressure at which the caudal artery pulse reappears during cuff deflation. This was repeated three times under standardised procedure to ensure reproducible results. Due to the hyperactive nature of the SHR strain, light anaesthesia (i.p. Zoletil® (tiletamine 15 mg/kg, zolazepam 15 mg/kg)) was necessary to obtain reproducible measurements. Anaesthesia was not required for the WKY rats. Measurements on WKY rats with and without light anaesthesia revealed a slight, non-significant drop in SBP with anaesthesia (112.05 ± 4.2 mmHg for conscious animals *versus* 106.18 ± 4.3 mmHg for anaesthetised animals ($n = 4$)).

2.3 Echocardiographic data

In vivo cardiac measurements were obtained by echocardiography, performed by qualified sonographers at the Prince Charles Hospital, Brisbane. The protocol used has been established and characterised by Brown *et al.* (2002) [16]. Rats were anaesthetised with an i.p. injection of Zoletil® (tiletamine 15 mg/kg, zolazepam 15 mg/kg) and Xylazine (10 mg/kg), which induced general anesthesia for 2 to 3 h. Images were obtained using a Hewlett-Packard Sonos 5500 and a 12-MHz neonatal transducer, with an image depth of 2 cm. Heart rate was recorded using a three-lead electrocardiogram attached to the front limbs and the hind right limb. Two-dimensional images and M-mode tracings were performed at the level of the papillary muscles and the aortic valves, while pulsed-wave Doppler analysis was used to measure mitral flow velocity at the tips of the mitral valve from the apical view. Structural parameters assessed included LV mass, relative (ventricular) wall thickness and diastolic and systolic LV internal diameters. Functional parameters included left ventricular ejection fraction (LVEF), which measures the percentage of blood ejected by the heart each beat and fractional shortening (FS), which is the change in LV diameter from diastole to systole.

2.4 Biochemical analysis

2.4.1 Tissue preparation and protein estimations

At the time of sacrifice, hearts were removed, weighed, and stored at –80°C. Samples were homogenised in four volumes of extraction buffer (50 mM Tris-Cl, pH 7.5; 2 mM EDTA) using an Ultra Turax homogeniser (Heidolph IKA-Werke GMBH KG, Staufen, Germany) and then centrifuged

at 3500 × *g* for 30 min to separate cellular debris. Aliquots of supernatant were stored at –80°C. Prior to use, thawed tissue extracts were centrifuged at 14 500 × *g* for 15 min. Protein estimations were performed using a BCA Protein Assay kit (Pierce, Rockford, USA), following the manufacturer's protocol and using BSA as standard. Protein concentrations were expressed as milligram protein *per* millilitre of tissue extract.

2.4.2 Gpx assay

GPx activity was measured spectrophotometrically using a method modified from Flohe and Gunzler (1984) and adapted to work in a 96-well microplate [20, 21]. An assay mix was prepared, containing: Potassium phosphate buffer (KPi), pH 7.4, 50 mM; EDTA, 5 mM; glutathione reductase, 1.0 U/mL; glutathione, 2 mmol/L; and NADPH, 0.3 mmol/L. Heart extracts (5 μL) were added to a 96-well flat bottom plate in triplicate. Assay mix (180 μL) was added to all wells and the reaction was initiated with 10 μL of 10 mM *tert*-butyl hydroperoxide. The rate of NADPH oxidation was recorded at 340 nm over 5 min using a Tecan Sunrise Absorbance Reader with Magellan Standard software (TECAN, Austria). The activity of GPx was determined as moles/min/mL of extract. To standardise the enzyme activity, ratios of enzyme activity to mg of extracted protein were determined (moles/min/mg protein).

2.4.3 Thx-Red assay

Thx-Red activity was measured spectrophotometrically using a method modified from Arner *et al.* (1999) [22]. An assay mix containing: KPi, pH 7.0, 50 mM; EDTA, 5 mM; NADPH, 0.66 mg/mL; and Insulin, 2.16 mg/mL, was prepared. The assay was set up in a 96-well flat bottom plate and 120 μL of assay mix was added to the required wells (four wells *per* sample). Heart extracts (5 μL) were then added to the wells. Thioredoxin (10 μL of 1.4 mg/mL) was added to duplicate wells of each sample, while assay mix (10 μL) was added to the remaining two wells of each sample to measure background absorbance. The plate was then incubated at 37°C for 20 min, the reaction was stopped by adding Dithiobis(2-nitrobenzoic acid) (DTNB) (50 μL of 0.4 mg/mL DTNB in 6 M guanidine hydrochloride) and the plate was incubated at room temperature for 10 min then read at 412 nm using a Tecan Sunrise Absorbance Reader with Magellan Standard software (TECAN, Austria). Thx-Red activity was standardised by calculating the enzyme activity *per* milligram protein (moles/min/mg protein).

2.4.4 Protein carbonyl ELISA

Protein carbonyls were measured as a marker of oxidative injury using a quantitative immunoassay as described by Buss and Winterbourn (2002) [23] and modified by Alamdari *et al.* (2005) [23, 24]. Tissue extracts were reacted with 2,4-dinitrophenylhydrazine and then non-specifically adsorbed to wells of an ELISA plate. An antibody raised against protein-conjugated 2,4-dinitrophenylhydrazine (Sigma, Sydney, Australia) was then used to probe the adsorbed protein. This was followed by second antibody conjugated with horseradish peroxidase (Molecular Probes, Eugene, OR, USA) for quantification. A standard curve was prepared by mixing varying proportions of oxidised (0–24% v/v) and reduced BSA at a constant protein concentration of 3 mg/mL [25]. Protein carbonyl levels were expressed as the percent of oxidised protein *per* microgram of protein.

2.5 Data analysis

Data was analysed using one-way analysis of variance followed by Newman–Kuels and Bonferroni *post hoc* tests for multiple comparisons when initial differences were detected. All statistical tests were performed using SPSS (version 12.0.1) and $p < 0.05$ was considered statistically significant. All data values are reported as mean ± SEM unless otherwise indicated.

3 Results

3.1 Physiological measurements

3.1.1 Body weight

All rats, irrespective of diet and strain, showed an overall decrease in body weight with age (Fig. 1). Although not significant, there did appear to be a difference between the SHR diet groups, with greater weight loss demonstrated in the Se-free group (Se-free: 13.3 ± 4.8%; normal Se: 3.9 ± 0.9%; and high Se: 9.2 ± 1.9% (Se-free *versus* normal Se, $p = 0.07$)).

3.1.2 SBP

At baseline, there was no difference in the SBP between the three diet groups for each rat strain (Fig. 2). Over the 7-month period, WKY rats that received normal and high Se showed a marginal increase in SBP, whereas WKY rats that received the Se-free diet demonstrated a significant increase in SBP by 90 wk (125.7 ± 2.7 mmHg for Se-free *versus* 116.6 ± 3.0 mmHg for normal Se and 114.6 ± 3.4 mmHg for high Se, $p < 0.05$).

For the SHRs, all three diet groups showed a dramatic rise in SBP from the mean baseline measurement of 176.8 ± 4.6 mmHg at 63 wk to the second mean measurement of 212.3 ± 3.5 mmHg at 70 wk. At 78 wk, the SBP for SHRs receiving high Se appeared to plateau

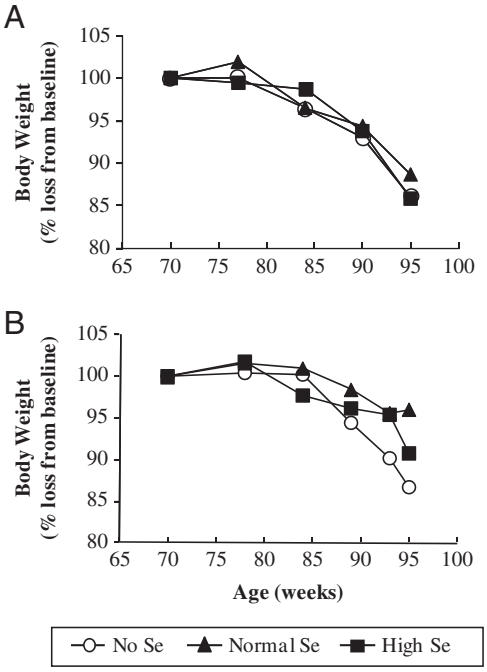

Figure 1. Changes in the body weight of (A) WKY rats and (B) SHRs over the 7-month dietary period. Values represent body weight as a percentage of baseline body weight. No significant difference was detected between the diet groups for either WKY or SHR.

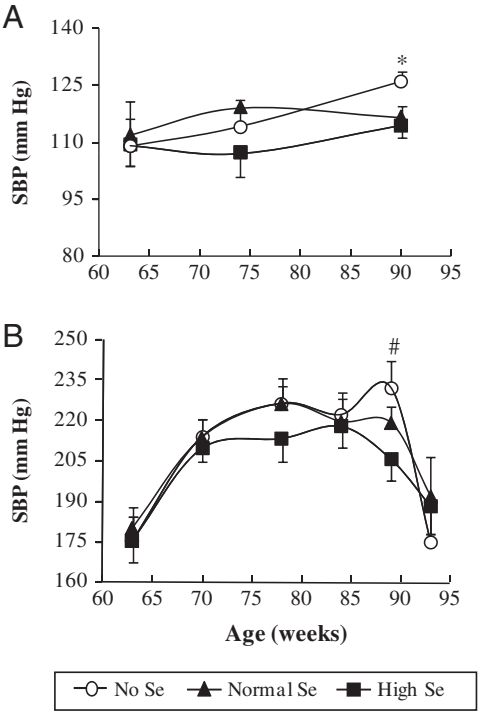

Figure 2. SBP measurements for (A) WKY rats and (B) SHRs recorded over the 7-month dietary period. Values represent mean (mmHg) ± SEM. At 89 (SHR) and 90 wk (WKY) of age, the following statistically significant differences were detected between the dietary groups: $^{*}p < 0.05$ (no Se *versus* both normal Se and high Se); $^{\sharp}$ $p < 0.05$ (no Se *versus* high Se).

(213.2 ± 8.5 mmHg), whereas the SBP for animals receiving Se-free or normal Se continued to rise (Se-free: 226.0 ± 6.8 mmHg; normal Se: 226.1 ± 9.0 mmHg). At 89 wk, the SBP for the Se-free group continued to rise to 231.0 ± 10.1 mmHg, while the measurements for the normal Se group began to plateau (219.1 ± 5.7 mmHg) and high Se group dropped slightly (205.5 ± 7.7 mmHg) (Se-free *versus* high Se, $p < 0.05$). At 93 wk of age, all SHRs showed a drop in SBP, although to different extents depending on diet. The largest drop was seen in animals that received the Se-free diet, decreasing from 231.0 ± 10.1 mmHg at 89 wk to 174.9 ± 15.0 mmHg. SHRs that received normal levels of Se showed a drop in SBP from 219.1 ± 5.7 mmHg to 192.0 ± 14.0 mmHg, while the least variation was observed in rats that received high levels of Se, with a drop from 205.5 ± 7.7 to 187.9 ± 10.0 mmHg.

3.1.3 Echocardiography

Echocardiographic assessment of WKY rats showed no detectable difference between the three diet groups for any of the LV structural or functional parameters (results not shown). Echocardiographic changes for the three SHR diet groups are shown in Fig. 3. All diet groups showed an overall increase in LV mass (Fig. 3A); however at 92 wk of age, this appeared to be slightly higher in the Se-free group (1.714 ± 0.100 g for Se-free *versus* 1.621 ± 0.134 g for normal Se and 1.496 ± 0.082 g for high Se). This was accompanied by thinning of the LV wall for all diet groups (relative wall thickness; Fig. 3D) and there was evidence of increased thinning in the Se-free group (0.397 ± 0.016 cm for Se-free *versus* 0.433 ± 0.026 cm for normal Se and 0.425 ± 0.014 cm for high Se). The diameter of the left ventricle, taken during both diastole (Fig. 3B) and systole (Fig. 3C), showed a progressive increase over time. Once again, at 92 wk of age, the observed increase in LV diameter was exaggerated in the Se-free group (diastole: 1.025 ± 0.037 cm for Se-free *versus* 0.928 ± 0.039 cm for normal Se and 0.958 ± 0.018 cm for high Se; systole: 0.611 ± 0.057 cm for Se-free *versus* 0.510 ± 0.039 cm for normal Se and 0.445 ± 0.028 cm for high Se (Se-free *versus* high Se, $p = 0.009$)).

All SHR diet groups showed a progressive decline in LV function, as evidenced by a decrease in LVEF (Fig. 3E) and FS (Fig. 3F). The decline in function was similar in the three groups; however at 92 wk there does appear to be a worsening in LV function in animals that received a Se-free diet (LVEF: 78.4 ± 3.7% for Se-free *versus* 83.8 ± 2.6 for normal Se and 85.4 ± 1.6% for high Se; FS: 40.9 ± 3.8% for

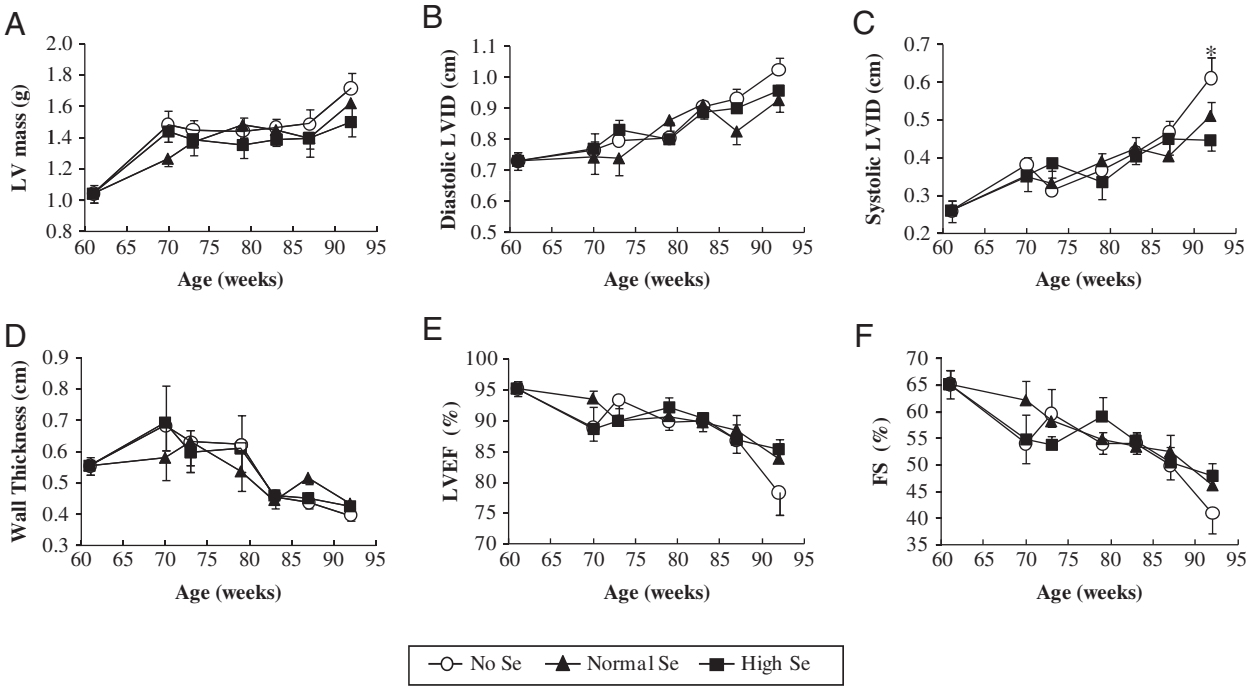

Figure 3. Echocardiographic assessment of LV structural and functional parameters for the SHR dietary groups recorded over the 7-month period. Parameters assessed included: (A) LV mass (grams); (B) diameter of LV during diastole (cm); (C) diameter of LV during systole (cm); (D) relative LV wall thickness (cm); (E) FS (%); and (F) LV ejection fraction (%). $^{*}p<0.05$ (no Se *versus* high Se).

Se-free *versus* 46.3 ± 2.8 for normal Se and 48.0 ± 2.4% for high Se).

3.1.4 Survival data

Survival data for both WKY and SHRs over the 7-month diet period is shown in Fig. 4. For the WKY rats, the lowest survival rate was seen in the Se-free group, with a total survival of 67% (four out of six). A survival rate of 83% (five out of six) and 100% (six out of six) was observed for WKY rats that received normal and high levels of Se, respectively. Of the deaths in the Se-free group, one animal died from respiratory complications following anaesthesia and one animal was euthanised for an unknown illness (weight loss, lethargy, and epistaxis). The death of the WKY in the normal Se group was also a result of respiratory complications following anaesthesia.

For the SHRs, the lowest survival rate was observed in the Se-free group, with a total survival of only 30% (three out of ten). A survival rate of 78% (seven out of nine) was observed in SHRs that received normal Se and a survival rate of 100% (nine out of nine) was observed in SHRs that received high Se. In the SHR rat, the observed mortality in the Se-free group was most likely due to the progression of hypertension-induced heart failure, with animals exhibiting classical signs such as weight loss, shortness of breath, and lethargy prior to either a natural death ($n = 3$) or euthanasia for animal welfare ($n = 4$).

3.2 Biochemical measurements

3.2.1 Antioxidant enzyme activity

Both rat strains showed an increase in cardiac GPx activity with increasing dietary Se (Fig. 5A). Animals fed the Se-free diet displayed extremely low levels of cardiac GPx, while the normal Se diet caused a substantial increase for both rat strains (approximately fivefold from Se-free). The high Se diet caused a further increase in the activity of cardiac GPx, although to different degrees depending on the rat strain. For the high Se group, SHRs demonstrated significantly higher activity levels of cardiac GPx compared with WKY rats (171.4 ± 9.0 mU/mg for SHR *versus* 126.4 ± 6.4 mU/mg for WKY, $p<0.001$). A similar trend was observed for cardiac Thx-Red activity (Fig. 5B), although once again strain differences were observed. SHRs showed a significant increase in Thx-Red activity from Se-free to normal Se (3.4-fold increase), and then a further significant increase with high Se (1.5-fold increase from normal Se).

3.2.2 Oxidative stress

As a measure of oxidative injury, heart tissue extracts were analysed for levels of protein carbonyls (Fig. 5C). The levels of protein carbonyls in the tissues from WKY rats were negligible in all experimental dietary groups. Cardiac

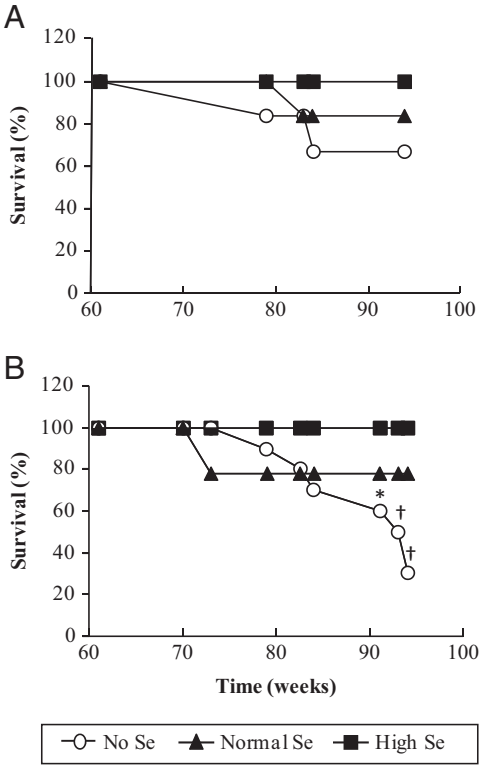

Figure 4. Survival data for (A) WKY rats and (B) SHRs over the 7-month dietary period. Values represent survival rate as a percentage of baseline. At initiation of diet the sample sizes were: no Se: WKY = 6 and SHR = 10; normal Se: WKY = 6 and SHR = 9; and high Se: WKY = 6 and SHR = 9. $^{*}p<0.05$ (no Se *versus* both normal Se and high Se); † $p<0.01$ (no Se *versus* both normal Se and high Se).

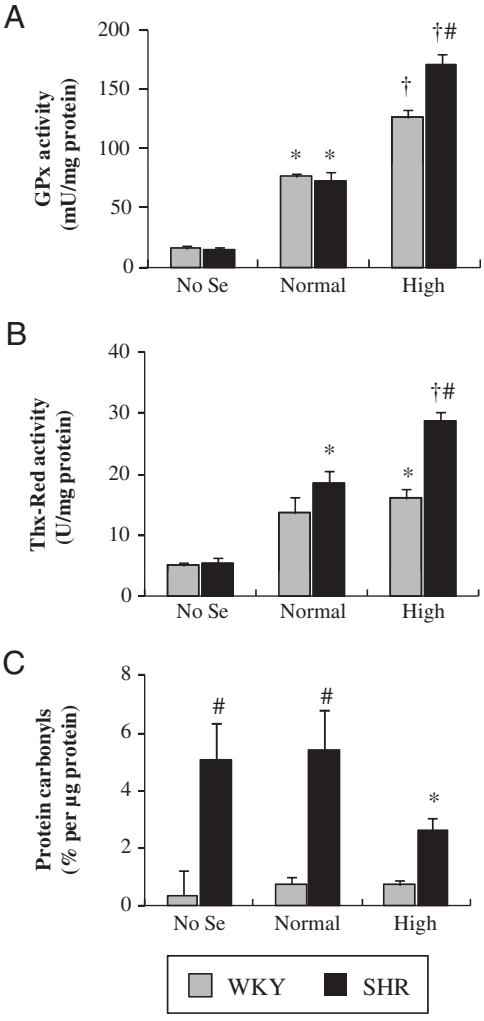

Figure 5. Measurement of antioxidant enzyme activity and oxidative injury in heart extracts from SHR and WKY rats on the three experimental diets. (A) GPx activity (mU/mg protein); (B) Thx-Red activity (U/mg protein); and (C) Levels of protein carbonyls (% *per* µg protein). U = moles/min. $^{*}p<0.05$ (*versus* the no Se diet group for the same rat strain); † $p<0.05$ (*versus* the normal Se diet group for the same rat strain); ♯ $p<0.05$ (SHR *versus* WKY for the same diet group). NB: Se analysis was performed on heart extracts; however the levels were too low for detection (<0.05 mg/kg tissue).

tissue from SHRs that received a Se-free diet showed the highest levels of oxidised proteins. SHRs that received a diet containing normal Se still showed a high level of heart tissue protein carbonyls, similar to the levels seen in SHRs receiving a Se-free diet, whereas a diet high in Se showed a reduction in oxidised protein levels. The reduction in the levels of heart protein carbonyls in the SHR rat on the high Se diet was found to be significant (*versus* no Se, $p<0.05$).

4 Discussion

The most striking finding of this study was the high mortality rate observed in SHRs fed a diet deficient in Se, with only 30% of Se-free SHRs surviving to study end (Fig. 4B). A diet containing normal (50 µg Se/kg feed) or high (1000 µg Se/kg feed) levels of Se seemed to delay the onset of severe CHF and subsequent death in the SHRs, with a survival rate of 78 and 100%, respectively. The observed mortality in the SHRs fed a Se-free diet was most certainly due to the progression of hypertension-induced heart failure, with animals exhibiting classical signs such as weight loss, dyspnoea, and lethargy as well as echocardiographic changes consistent with heart failure, prior to either a natural death or euthanasia.

The development of hypertension was also influenced by dietary Se, with chronic Se deficiency associated with higher SBP measurements in both rat strains (Figs. 2A and B). As the SHRs progressed to heart failure, a dramatic drop in blood pressure was observed, most likely as a result of impaired compensatory mechanisms in the failing heart. More specifically, in end-stage heart failure, cardiac output cannot be maintained due to decreased contractility of the

left ventricle, resulting in hypotension. This suggests that either dietary Se enhances cardiac compensatory mechanisms in end-stage heart failure or that the animals receiving normal or high levels of Se had not yet progressed to end-stage heart failure. The latter suggestion is more likely, especially when interpreted along with the survival data.

Cardiac changes in both rat strains were followed using echocardiography. Changes attributable to the normal ageing process were observed in the normotensive WKY rats and these changes were not influenced by Se deficiency or supplementation (results not shown). Aged SHRs showed cardiac changes consistent with the development of hypertension-induced cardiac hypertrophy and dysfunction (Figs. 3A–F). Cardiac hypertrophy was evidenced by a marked increase in LV mass, with a concomitant dilation of the LV chamber and thinning of the LV wall. Impairment of cardiac function was also evident, indicated by a decline in LVEF and FS values. Chronic Se deficiency in SHRs was associated with a more pronounced cardiac hypertrophy and a worsening of systolic functional parameters when compared with SHRs that received normal or high levels of dietary Se. It should be noted that this dietary group had a substantially reduced sample size due to high mortality, which inevitably reduced the power of the study so statistical significance was not proven although an obvious trend was observed. Due to this high mortality and loss of power these data should be considered as preliminary observations and need to be reinforced by a larger study.

At a biochemical level, analysis of cardiac tissues from SHR and WKY rats revealed that dietary Se was able to effectively modulate the activity of the selenoenzymes GPx and Thx-Red. This provides a simple and applicable model of modulating endogenous protein expression and activity *in vivo* without the need for artificial regulation of gene expression. We have used this to generate ''antioxidant depleted'' animals, which we have used in a variety of studies [8, 9]. Our recent work with cardiac patients also suggests that Se supplementation, in association with other antioxidants, may prove beneficial in preparing patients for cardiac surgery [26].

Human hypertension is thought to be a combination of environmental triggers and a genetic predisposition [15]. The pathogenesis is variable, although it usually involves either excessive vasoconstriction or reduced vasodilation [15]. In normal physiology, vasodilation is controlled by three main factors: prostacyclin, nitric oxide, and endothelium-derived hyperpolarising factor [27]. Several studies have established a role for oxidative stress in the development of hypertension, through interference with the regulation of vascular tone. Reactive oxygen species (ROS) have been found to irreversibly inactivate prostacyclin synthase [28] and decrease nitric oxide bioavailability [29], impairing vasodilation. Furthermore, ROS directly oxidise arachidonic acid, forming isoprostanes, which have been shown to exert potent vasoconstrictor activity [30–32]. Increased levels of ROS and/or oxidative stress markers, as well as reduced levels of antioxidants, have been demonstrated in hypertensive patients and the administration of antioxidants in animal models of hypertension have been shown to reduce or abrogate the development of a hypertensive state [1, 2, 30, 33–38].

Antioxidants have also been shown to reduce or attenuate the progression from compensated to decompensated hypertrophy, further supporting the role of oxidative stress in the pathophysiology of myocardial remodeling during heart failure [6, 39, 40]. GPx is considered of primary importance in the cellular defense against oxidative stress and, *in vitro*, has been demonstrated to have greater protective effects than superoxide dismutase or catalase [41]. Furthermore, a prospective study of cardiovascular risk deemed a decrease in GPx activity as one of the strongest predictors for an increased risk of cardiovascular disease, whereas no such association was revealed for superoxide dismutase activity [42]. A decline in myocardial GPx has been demonstrated specifically during the progression from compensated to decompensated hypertrophy [43] and direct evidence of the protective effect of GPx in myocardial remodeling has recently been established using a GPx transgenic mouse [44]. Overexpression of GPx inhibited matrix metalloproteinase activation, decreased interstitial fibrosis, and reduced cardiac hypertrophy and dysfunction post-myocardial infarction, as well as improving survival rate. These findings clearly implicate GPx in the pathogenesis of cardiac remodelling and the subsequent progression to heart failure. However, it should be noted that Se depletion may result in reduced activity of other important selenoproteins, some of which have been implicated in cardiomyocyte development and function [46].

In conclusion, dietary Se was found to influence the development of both genetic and aged-related hypertension as seen in the SHR and WKY rats, respectively. Dietary Se neither affected age-related cardiac changes in the WKY nor the mortality rates. However in the SHR, increased Se intake was clearly associated with an increase in selenoantioxidant enzyme activity and a decrease in cardiac oxidative injury. Dietary Se intake was further correlated with a reduction in disease severity and mortality in the SHR. These observations emphasise the importance of maintaining adequate dietary Se in individuals at higher risk of developing CHF. This may be particularly relevant in ageing populations, hypertensive individuals, or in countries where the Se content of food is falling.

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The authors have declared no conflict of interest.

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