Effects of subacute dietary salt intake and acute volume expansion on diastolic function in young normotensive individuals

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Aims
Chronic excess salt intake may have blood pressure-independent adverse effects on the heart such as myocardial hypertrophy and fibrosis. Effects of subacute sodium loading with excess dietary salt on diastolic function in normotensive individuals have been conflicting and the mechanisms are poorly understood.

Methods and results
Thirteen healthy normotensive subjects (age 24 ± 4 years) entered a 2-week crossover study with 1 week of a low-salt diet, 10 mEq/day and 1 week of a high-salt diet, 200 mEq/day. At the end of each study week, left ventricular dimensions, systolic, and diastolic function were assessed with echocardiography before and after 2 L of normal saline infusion. One week of high-salt and low-salt diets did not lead to differences in echocardiographic parameters of systolic or diastolic function, even after rapid volume expansion with saline infusion. The peak early diastolic strain rate (SR) increased after volume loading both after completion of low-salt (1.62 ± 0.23/s vs. 1.82 ± 0.14/s, P < 0.05) and high-salt diets (1.67 ± 0.16/s vs. 1.86 ± 0.22/s, P < 0.05). There was a positive correlation between the peak early diastolic SR and the cardiac index (r = 0.52, P = 0.017).

Conclusion
In healthy normotensive individuals, subacute excess dietary sodium intake does not affect diastolic function. The peak early diastolic SR, similar to other mitral Doppler and tissue Doppler parameters of diastolic function, appears to be strongly dependent on pre-load.

Keywords
Diastolic function • Diastolic strain rate • Sodium

Introduction
Chronic excess intake of dietary salt can cause structural and functional damage to the heart. In rats, chronic excess dietary salt intake for 4 weeks has been reported to induce left ventricular (LV) hypertrophy and diastolic dysfunction. Similarly, chronic salt excess in humans may ultimately contribute to diastolic dysfunction and heart failure. The effect of shorter-term excess dietary salt intake on diastolic function, however, is controversial. Tzemos et al. reported that a 5-day period of salt loading impaired LV relaxation in healthy normotensive volunteers. In contrast, Williams et al. noted that increased dietary sodium for 1 week in normotensive subjects led to changes in indexes of LV diastolic function that may suggest improved diastolic function in normotensive subjects. The latter investigators suggested that the suppression of the renal artery aldosterone system may contribute to improvements in diastolic function, as angiotensin II was reported to impair indexes of early diastolic relaxation.

In the evaluation of diastolic function, acute isotonic volume expansion has been utilized to unmask abnormalities of cardiovascular responses. In particular, acute volume loading with saline infusion can unmask potential intrinsic myocardial relaxation abnormalities in patients with untreated mild-to-moderate hypertension. The assessment of diastolic function using mitral Doppler flow velocities depends on the loading conditions.
velocities by tissue Doppler imaging (TDI) may be less dependent on load in the evaluation of diastolic function. For example, LV filling pressures appear to have a minimal effect on the early diastolic mitral annular velocity $E'$ in the presence of impaired relaxation. However, in subjects with normal relaxation, $E'$ has been shown to increase with pre-load.\(^{10,11}\)

In recent years, the early diastolic strain rate (SR) has been suggested to be a reliable indicator of myocardial relaxation\(^{12–14}\) and of subtle LV diastolic dysfunction.\(^{15}\) Studies have shown that the peak early diastolic SR is a sensitive indicator of diastolic dysfunction in coronary artery disease, mitral regurgitation, and hypertrophy cardiomyopathy.\(^{12,16–18}\) Although there is robust evidence about the load dependency of systolic strain and recent studies suggesting that systolic SR is also load dependent,\(^{19–23}\) there is limited data on the effect of acute changes in loading conditions on diastolic SR in humans.

The aim of the study was to investigate the impact of short-term excess dietary salt intake on cardiac function in young healthy volunteers, in particular diastolic function including the diastolic SR. We hypothesized that these novel measures and an acute volume load would be able to unmask potential diastolic dysfunction in subjects who had been pre-treated with a high-salt diet.

**Methods**

**Patient selection**

The study was approved by the institutional board review and all volunteers provided written informed consent. Volunteers aged between 18 and 40 years old were recruited. Exclusion criteria included the presence of hypertension or other cardiac, pulmonary, rheumatologic, renal, or any disease that could affect LV diastolic or systolic function.

**Protocol**

Each volunteer was maintained on the low-salt diet for 1 week and on the high-salt diet for 1 week. The order of the diet was randomized and a washout period of at least 2 weeks between the diets was included. The low-salt diet contained 10 mEq/day of sodium and the high-salt diet contained 200 mEq/day of sodium.\(^5\) All meals were prepared and provided to subjects for the week. Subjects were expected to consume all food as provided, with no substitutions or additions. On Days 5–6 of the assigned diet, subjects performed a 24-h urine collection to confirm adherence to the diet. Prior to the scheduled protocol visit, subjects were excluded if the 24-h sodium urinary excretion was $>20$ mEq on the low-salt diet or $<180$ mEq on the high-salt diet. All the subjects included in this study met the requirements and confirmed dietary compliance.

After 1 week, each of a low-salt and high-salt diet, each participant underwent a scheduled visit and was admitted to the Clinical Research Unit overnight. In the morning after an overnight fast, a two-dimensional echocardiogram was first recorded. Isotonic saline was then infused at 0.25 mL/kg/min for 2 h as previously described.\(^{24}\) Heart rate and blood pressure were measured at 15-min intervals by a standard sphygmomanometer. Subjects were asked to void every 30 min. Urine was collected for the assessment of the sodium excretion rate. All urine output was replaced with distilled water by mouth, in addition to 40 mL/h to account for insensible losses. At the end of the saline infusion, another echocardiographic study was obtained.

**Echocardiographic examination**

All echocardiographic examinations were performed using the Vivid-7 (GE Healthcare, Milwaukee, WI, USA). Two-dimensional, Doppler, and colour TDI were obtained from standard parasternal, apical, and subcostal views. All data were stored digitally for post-study offline data analysis. Interpretation of the echocardiograms was blinded to the diet or saline loading conditions of each subject. The LV volume and the ejection fraction were calculated from the apical four- and two-chamber views using the modified biplane Simpson’s method. Mitrail E and A waves were measured from the apical four-chamber view by placing the sample volume at the leaflet tips of the open mitral valve. Spectral pulsed tissue Doppler velocity data were acquired from an optimal measuring position at the basal segment of the septal and lateral walls of the LV using a 2 mm sample volume. Myocardial longitudinal velocities during three consecutive cardiac cycles were recorded while care was taken to keep the data sampling point placed within the ventricular myocardium and the angle of beam as parallel as possible to the long axis of the myocardial movements during each cardiac cycle. Colour M-mode Doppler of LV mitral inflow in early diastole was obtained from the apical four-chamber view. The ratio of the mitral inflow to mitral annular peak early diastolic velocity ($E/E'$) was calculated. Left atrial volumes were calculated using the biplane Simpson’s method.

Speckle-tracking imaging and strain analysis were undertaken as previously described.\(^{25,26}\) Images were acquired with the frame rate between 80 and 100 frames/s. Myocardial systolic and diastolic longitudinal strain and SR were measured offline using speckle-tracking analysis on the apical views (EchoPAC, version 8, GE Healthcare). Peak global strain was calculated as the average strain in the basal, mid-, and apical LV as measured in the apical four-, two-, and three-chamber views. Peak systolic strain ($S$), peak systolic SR ($SR_S$), peak early diastolic strain ($SR_E$), and peak late diastolic SR ($SR_A$) were recorded in each view for each of the six segments pre-defined by the two-dimensional strain algorithm and averaged. Global peak systolic and diastolic longitudinal strain was calculated by averaging the values of peak systolic and diastolic strain of all 18 segments. The measurements were reported as the average of three consecutive cardiac cycles.

**Intra-observer and inter-observer variability**

For the evaluation of the intra-observer and inter-observer measurement variability, a group of randomly selected recordings were analysed by two independent investigators (G.S.M. and H.S.) who were unaware of each other’s measurements and of the diet content or study time point. To measure intra-observer variability, a single observer (G.S.M) repeated the measurements on the same loops several weeks after the first measurement set. Intra- and inter-observer reproducibility were assessed by intra-class correlation coefficient. Repeat analysis of TDI velocities data showed 0.96 (95% CI: 0.91–0.98) for intra-observer variability and 0.93 (95% CI: 0.86–0.97) for inter-observer variability. For peak early diastolic SR, the intra-observer variability was 0.97 (95% CI: 0.93–0.99) and inter-observer variability was 0.89 (95% CI: 0.76–0.95).

**Statistical analysis**

Statistical analysis was performed using the statistics software SPSS (SPSS, Inc., Chicago). Data are presented as mean ± standard deviation unless otherwise specified and a P-value <0.05 was considered significant.

The effect of dietary sodium intake and acute volume loading were $a$ priori considered to be separate experiments. The effect of dietary sodium intake was first analysed using paired t-tests. The independent
were not different. There was no significant difference in
systolic or diastolic blood pressure after low-salt or
high-salt diets. However, there was a trend towards a lower heart
rate after the high-salt diet compared with the low-salt diet. There was no
difference in systolic or diastolic blood pressure after low-salt or
high-salt diets. However, there was a trend towards a lower heart
rate after the high-salt diet compared with the low-salt diet. There was no

Results

Study population and baseline characteristics

Thirteen subjects (male = 12, female = 1), with a mean age of
24 ± 4 years, completed the full protocol. The mean height of
the subjects was 1.8 ± 0.1 m, with a mean body mass index
of 23.7 ± 1.4 kg/m².

Effect of dietary sodium and saline infusion on body weight and
haemodynamics

The effects of dietary sodium and saline infusion on body weight and
haemodynamics are summarized in Table 1. The total body
weight of the subjects was higher at the completion of the high-salt
diet than at the completion of the low-salt diet. There was no
difference in systolic or diastolic blood pressure after low-salt or
high-salt diet. However, there was a trend towards a lower heart
rate after the high-salt diet compared with the low-salt diet
(P = 0.1). Saline infusion did not significantly modify the heart
rate, systolic, or diastolic blood pressure.

Effect of dietary sodium on
echocardiographic parameters of systolic
and diastolic function

The comparison of echocardiographic measurements for subjects
on low-salt vs. high-salt diet is outlined in Tables 2 and 3. The
stroke volume was higher at the completion of the high-salt diet
than after the low-salt diet, but the cardiac output was not significa-
cantly different. The LV end-diastolic and end-systolic volumes
were not different. There was no significant difference in
echocardiographic measures of systolic function, including LV ejection
fraction, tissue Doppler systolic mitral annular velocities, peak
systolic longitudinal strain, and SR. Similarly, there was no differ-
ence in echocardiographic measures of diastolic function, including
mitral inflow parameters, diastolic mitral annular velocities, mitral

<table>
<thead>
<tr>
<th>Table 1</th>
<th>Body weight, blood pressure, and heart rate of subjects on a low-salt or high-salt diet, before and after saline infusion</th>
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<tbody>
<tr>
<td></td>
<td>Low salt</td>
</tr>
<tr>
<td></td>
<td>Pre-saline</td>
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<tr>
<td>Weight (kg)</td>
<td>74.0 ± 7.9</td>
</tr>
<tr>
<td>SBP (mmHg)</td>
<td>114 ± 5</td>
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<tr>
<td>DBP (mmHg)</td>
<td>68 ± 7</td>
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<tr>
<td>HR (bpm)</td>
<td>60 ± 10</td>
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DBP, diastolic blood pressure; HR, heart rate; SBP, systolic blood pressure.
*P < 0.05 compared with pre-saline value of the low-salt diet.

<table>
<thead>
<tr>
<th>Table 2</th>
<th>Echocardiographic parameters of healthy volunteers after completion of low-salt and high-salt diets</th>
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<tbody>
<tr>
<td></td>
<td>Low-salt</td>
</tr>
<tr>
<td>IVS (cm)</td>
<td>0.90 ± 0.16</td>
</tr>
<tr>
<td>PWVt (cm)</td>
<td>0.91 ± 0.13</td>
</tr>
<tr>
<td>LVEDV (mL)</td>
<td>115 ± 27</td>
</tr>
<tr>
<td>LVESV (mL)</td>
<td>41 ± 13</td>
</tr>
<tr>
<td>LVEF (%)</td>
<td>64.6 ± 5.2</td>
</tr>
<tr>
<td>SV (mL)</td>
<td>74 ± 16</td>
</tr>
<tr>
<td>CO (L/min)</td>
<td>4.2 ± 1.1</td>
</tr>
<tr>
<td>E (cm/s)</td>
<td>76.5 ± 19.0</td>
</tr>
<tr>
<td>A (cm/s)</td>
<td>41.1 ± 12.5</td>
</tr>
<tr>
<td>S’lat (cm/s)</td>
<td>10.9 ± 1.8</td>
</tr>
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<td>E’lat (cm/s)</td>
<td>20.1 ± 1.7</td>
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<tr>
<td>A’lat (cm/s)</td>
<td>6.9 ± 1.5</td>
</tr>
<tr>
<td>E/A</td>
<td>1.90 ± 0.33</td>
</tr>
<tr>
<td>E/E’lat</td>
<td>3.89 ± 1.06</td>
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<tr>
<td>LAVI (mL/m²)</td>
<td>23.3 ± 4.1</td>
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<tr>
<td>DT (ms)</td>
<td>192 ± 32</td>
</tr>
<tr>
<td>IVRT (ms)</td>
<td>85 ± 15</td>
</tr>
<tr>
<td>Vp (cm/s)</td>
<td>62 ± 13</td>
</tr>
<tr>
<td>Strain peak S (%)</td>
<td>−17.9 ± 1.2</td>
</tr>
<tr>
<td>SR peak S (ls)</td>
<td>−1.07 ± 0.11</td>
</tr>
<tr>
<td>SR peak E (ls)</td>
<td>1.62 ± 0.23</td>
</tr>
<tr>
<td>SR peak A (ls)</td>
<td>0.61 ± 0.10</td>
</tr>
</tbody>
</table>

CO, cardiac output; DT, deceleration time; IVRT, intra-ventricular relaxation time; IVS, inter-ventricular septum; LAVI, left atrial volume index; LVEDV, left ventricular end-diastolic volume; LVESV, left ventricular end-systolic volume; PWVt, posterior wall thickness; SR, strain rate; SV, stroke volume; Vp, flow propagation velocity.
*P < 0.05 compared with the pre-saline value of the low-salt diet.
flow propagation velocity, and peak diastolic longitudinal SR. The left atrial volume was unchanged.

Effect of saline infusion in subjects on low-salt and high-salt diet

The LV end-diastolic volume increased after volume expansion with saline infusion, whereas LV end-systolic volume was unchanged. Both the stroke volume and the cardiac output increased. An increase in LV ejection fraction and systolic peak tissue velocity by TDI (S') was noted with volume increase. There was a significant increase in longitudinal LV systolic strain and SR after saline infusion. The above effects were similar for subjects on both the low-salt and high-salt diet conditions. In patients with L-wave, other diastolic parameters in the subjects with L-wave compared with subjects without L-wave.

Saline infusion induced an increase in early diastolic (E) and late diastolic (A) mitral inflow velocities after both diets. The E/E' ratio increased similarly in both groups. No significant change was observed in late diastolic peak tissue velocity (A'), deceleration time, isovolumic relaxation time, or colour flow propagation velocity with volume change. Left atrial volume increased as a result of saline administration in both groups. We observed a prominent mid-diastolic wave, also described as L-wave, among all the studies in three subjects. These subjects were all bradycardic with the heart rate <60 bpm at the time of study. In one additional subject, the L-wave was only present after the high-salt diet; it is noteworthy that this subject was very bradycardic (40–45 bpm) at the time of the study. The L-wave did not change in amplitude with loading conditions. Corresponding L'-wave was also not apparent in the tissue Doppler recordings. Although the number of subjects was very small, there was no obvious difference in other diastolic parameters in the subjects with L-wave compared with subjects without L-wave.

After saline infusion, peak early diastolic longitudinal SR increased in both the high-salt and the low-salt diet conditions. Individual changes in the peak early diastolic SR (SR E) after volume loading are presented in Figure 1. There was a positive correlation between the cardiac index and the peak early diastolic SR (SR E) \( r = 0.52, P = 0.017 \), as shown in Figure 2. The peak late diastolic SR (SR A) did not change after saline infusion.

Discussion

In the present study, 1 week of the high-salt diet resulted in a small increase in stroke volume, but did not have any detectable effect on cardiac function, in particular diastolic function. Acute volume loading did not unmask any diastolic abnormalities after the
high-salt diet. The acute increase in pre-load from volume loading was associated with an augmentation of diastolic SR, underlining the load dependency of this parameter.

**Effect of dietary salt intake**

One week of high-salt feeding in healthy volunteers was accompanied by an increase in total body weight compared with low-salt feeding. This increase in total body weight was not associated with a significant increase in LV end-diastolic dimensions or cardiac output. Thus, the increase in total body weight may predominantly reflect an expansion of extracellular volume rather than an increase in intra-vascular volume. This finding is in accordance with results from previous investigators.\(^27,28\) Although a small increase in stroke volume was also noted, this increase did not result in a significant augmentation of cardiac output as the heart rate tended to be lower after the high-salt diet, a finding previously described by others.\(^29\) There was no difference in blood pressure induced by this short-term high-salt feeding.

There were no significant differences in Doppler or TDI parameters of systolic or diastolic function between high-salt and low-salt diets. Similarly, no differences in the early or late peak diastolic SR were observed for subjects on high-salt and low-salt diets. Thus, there are no differences between any diastolic echocardiographic indexes after 1 week of high-salt or low-salt feeding. Our findings do not agree with a previous study that 1 week of high dietary sodium intake impairs myocardial relaxation\(^4\) despite using a more extensive assessment of diastolic function.

**Figure 1** Changes in the peak early diastolic SR after volume loading with saline infusion in subjects on low-salt and high-salt diet.

**Figure 2** Relationship between the peak early diastolic SR and the cardiac index in subjects on low-salt and high-salt diet.
Effect of volume expansion by saline administration

Saline infusion led to an increase in LV end-diastolic volume and a relatively unchanged LV end-systolic volume, resulting in an increase in stroke volume and EF both after the low-salt and the high-salt diet. This augmentation of the stroke volume and the cardiac output is consistent with an increase in pre-load. Saline infusion also induced an increase in E and A velocities, most likely attributable to the pre-load increase. Although E and A are also affected by afterload, the lack of the effect of saline infusion on systolic blood pressure during volume expansion makes it unlikely that afterload was increased. Similarly, the increase in E' and E/E' observed after saline infusion is most likely due to the pre-load augmentation in these healthy individuals. Of note, no significant increase in E' was noted with saline infusion after high-salt feeding; however, the absolute value of E' after saline infusion was similar in both groups, arguing against a different diastolic function in high-salt and low-salt diets.

There were no significant changes in the mitral annular late diastolic wave (A') induced by an acute increase in pre-load. This lack of change with loading variations has been observed by others and may reflect the fact that the atrial contraction of normally functioning atrium appears to be less dependent on loading conditions than other diastolic parameters. The atrium has an important booster pump function due to its capacity for pre-systolic contraction. It has been previously demonstrated that the contribution of atrial contraction to filling depends on numerous factors, such as atrial contractility, pre-load, afterload, heart rate, and autonomic tones among others. The complexity of these factors in affecting the atrial contraction likely makes A' less susceptible to change in pre-load conditions.

A mid-diastolic wave, also described as L-wave, was observed in three subjects in each of their studies and in one subject after saline infusion is most likely due to the pre-load augmentation in these healthy individuals. Of note, no significant increase in E' was noted with saline infusion after high-salt feeding; however, the absolute value of E' after saline infusion was similar in both groups, arguing against a different diastolic function in high-salt and low-salt diets.

Important to consider the pre-load when analysing all diastolic parameters including the SR.

Limitations

Some limitations of the study should be noted. The small number of volunteers may mask an effect of the dietary sodium on diastole. However, the impairment in diastole was reported by others on a similar number of subjects. Our conclusions are also limited to young healthy individuals after a short duration of high-salt and low-salt diets. Although the amount of sodium in the high-salt diet is moderate, it has been employed in other studies to detect the cardiac effects of salt. Another limitation of this study is that its findings are limited to healthy normotensive individuals and the results may not be extrapolated to older hypertensive subjects with more prevalent diastolic dysfunction.

Conclusions

In healthy normotensive subjects, 1 week of high dietary sodium intake does not affect diastolic function, before and after acute volume loading. The acute volume load, however, increases all echocardiographic parameters of diastolic function, including the early diastolic SR, underlining the pre-load dependency of echocardiographic diastolic assessment. Changes in pre-load should be considered when interpreting the results of the diastolic SR in healthy hearts.

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References


