Nitric oxide (NO) is an important signalling molecule in the regulation of vascular resistance and has been implicated in the aetiology of hypertension. Hypertension is associated with decreased vagal tone, in patients with established hypertension, and in animal models of hypertension such as the spontaneously hypertensive rat (SHR). Hypertension is also associated with down regulation of guanylate cyclase (GC) in the aorta from SHRs, and this precedes the onset of hypertension. In the heart, the NO/guanylate cyclase/cyclic GMP dependent pathway enhances the negative chronotropic effect of vagal nerve stimulation. In addition, NO also facilitates the release of atrial natriuretic factor (ANF) by a pre-synaptic mechanism.

**Aim of the study**

The aim of this study was to establish whether the cholinergic regulation of heart rate is attenuated in hypertensive rats (SHRs) compared to normotensive (WKY) controls, and to establish if this is associated with reduced expression of guanylate cyclase.

**Methods**

32-40g male SHRs (n=20) and WKYs (n=26), 16-20 weeks old were used for the study.

A double aortic / right vagal preparation was dissected free, placed in an organ bath containing oxygenated Tyrode's solution (37°C) and connected to an isometric force transducer. Heart rate was triggered from contraction. The change in heart rate with vagal nerve stimulation for 30s or bath applied carbamylcholine (CCh, 100-500nmol/L) was measured. Drugs were added to the preparation after control protocols were completed.

Tissue samples (left ventricle, right atrium and aorta) from SHR and WKY rats were used for Western blot analysis. The primary antibody was polyclonal rabbit anti-sGC α1 subunit 82 kDa and rabbit anti-sGC α1 subunit 70 kDa.

**Protocols**

1. The heart rate response to vagal nerve stimulation (2Hz, 5Hz, 10Hz, 10-15V, 12ms duration for 25 seconds at 1 minute intervals) in SHR was compared with WKY rats. The order of the stimulations was randomised.

2. The heart rate response to bath applied CCh (100, 300, 500nmol/L) in SHR was compared with WKY rats.

3. The effect of the NO donor, SNP (2mmol/L), on the HR response to vagal nerve stimulation (at 5 and 10Hz) was compared in SHR and WKY rats. There was no significant difference in baseline heart rate (WKY, average = 224±7 bpm; SHR, average = 245±5 bpm).

4. The effect of SNP was studied on the heart rate response to vagal nerve stimulation (at 2Hz, 5Hz, 10Hz and 10-15V, 12ms duration for 25 seconds at 1 minute intervals) in SHR and WKY rats.

**Conclusions**

There is an impaired cholinergic response to peripheral vagal nerve stimulation in hypertensive (SHR) rats compared with normotensive (WKY) rats. This may result from altered pre-synaptic signalling, since the heart rate response to bath applied CCh was not attenuated in SHRs compared to WKYs.

This impairment is associated with down regulation of the α1 subunit of guanylate cyclase in the SHR. However, the mechanism by which hypertension interferes with guanylate cyclase expression is not yet known.

The reduced vagal tone demonstrated in this study in the hypertensive rat may contribute to the overall increased risk of sudden cardiac death in hypertensive patients since low vagal tone is a negative prognostic indicator of clinical outcome.

**Acknowledgements**

The authors thank Dr Simon Golding for his assistance with the molecular biology.

**References**


**Figure 1**

Nitric oxide (NO) is an important signalling molecule in the regulation of vascular resistance and has been implicated in the aetiology of hypertension. Hypertension is associated with decreased vagal tone, in patients with established hypertension, and in animal models of hypertension such as the spontaneously hypertensive rat (SHR). Hypertension is also associated with down regulation of guanylate cyclase (GC) in the aorta from SHRs, and this precedes the onset of hypertension. In the heart, the NO/guanylate cyclase/cyclic GMP dependent pathway enhances the negative chronotropic effect of vagal nerve stimulation. In addition, NO also facilitates the release of atrial natriuretic factor (ANF) by a pre-synaptic mechanism.

**Figure 2**

Aim of the study: The aim of this study was to establish whether the cholinergic regulation of heart rate is attenuated in hypertensive rats (SHRs) compared to normotensive (WKY) controls, and to establish if this is associated with reduced expression of guanylate cyclase.

**Figure 3**

Methods: 32-40g male SHRs (n=20) and WKYs (n=26), 16-20 weeks old were used for the study. A double aortic / right vagal preparation was dissected free, placed in an organ bath containing oxygenated Tyrode's solution (37°C) and connected to an isometric force transducer. Heart rate was triggered from contraction. The change in heart rate with vagal nerve stimulation for 30s or bath applied carbamylcholine (CCh, 100-500nmol/L) was measured. Drugs were added to the preparation after control protocols were completed.

Protocols: 1. The heart rate response to vagal nerve stimulation (2Hz, 5Hz, 10Hz, 10-15V, 12ms duration for 25 seconds at 1 minute intervals) in SHR was compared with WKY rats. The order of the stimulations was randomised.

2. The heart rate response to bath applied CCh (100, 300, 500nmol/L) in SHR was compared with WKY rats.

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4. The effect of SNP was studied on the heart rate response to vagal nerve stimulation (at 2Hz, 5Hz, 10Hz and 10-15V, 12ms duration for 25 seconds at 1 minute intervals) in SHR and WKY rats.

**Figure 4**

Conclusions: There is an impaired cholinergic response to peripheral vagal nerve stimulation in hypertensive (SHR) rats compared with normotensive (WKY) rats. This may result from altered pre-synaptic signalling, since the heart rate response to bath applied CCh was not attenuated in SHRs compared to WKYs. This impairment is associated with down regulation of the α1 subunit of guanylate cyclase in the SHR. However, the mechanism by which hypertension interferes with guanylate cyclase expression is not yet known.

The reduced vagal tone demonstrated in this study in the hypertensive rat may contribute to the overall increased risk of sudden cardiac death in hypertensive patients since low vagal tone is a negative prognostic indicator of clinical outcome.