Statins and the autonomic nervous system

Philip J. MILLAR* and John S. FLORAS*

*University Health Network and Mount Sinai Hospital Division of Cardiology, University of Toronto, Toronto, Ontario, Canada

Abstract
Statins (3-hydroxy-3-methylglutaryl-CoA reductase inhibitors) reduce plasma cholesterol and improve endothelium-dependent vasodilation, inflammation and oxidative stress. A ‘pleiotropic’ property of statins receiving less attention is their effect on the autonomic nervous system. Increased central sympathetic outflow and diminished cardiac vagal tone are disturbances characteristic of a range of cardiovascular conditions for which statins are now prescribed routinely to reduce cardiovascular events: following myocardial infarction, and in hypertension, chronic kidney disease, heart failure and diabetes. The purpose of the present review is to synthesize contemporary evidence that statins can improve autonomic circulatory regulation. In experimental preparations, high-dose lipophilic statins have been shown to reduce adrenergic outflow by attenuating oxidative stress in central brain regions involved in sympathetic and parasympathetic discharge induction and modulation. In patients with hypertension, chronic kidney disease and heart failure, lipophilic statins, such as simvastatin or atorvastatin, have been shown to reduce MNSA (muscle sympathetic nerve activity) by 12–30%. Reports concerning the effect of statin therapy on HRV (heart rate variability) are less consistent. Because of their implications for BP (blood pressure) control, insulin sensitivity, arrhythmogenesis and sudden cardiac death, these autonomic nervous system actions should be considered additional mechanisms by which statins lower cardiovascular risk.

Key words: blood pressure, nitric oxide, oxidative stress, sympathetic nervous system, statin

INTRODUCTION
Statins or HMG-CoA (3-hydroxy-3-methylglutaryl-CoA) reductase inhibitors act competitively on the mevalonate pathway to reduce endogenous production of cholesterol. Depending upon the dose and type of statin prescribed, reductions in total cholesterol between 15 and 40% and in LDL (low-density lipoprotein)-cholesterol between 20 and 55% can be achieved [1]. Large-scale trials, whether alone or integrated into meta-analyses, have demonstrated that statins reduce cardiovascular morbidity and mortality in high-risk populations [2–6] and the risk of major vascular events in low-risk populations [7]. As a consequence, statins have become one of the most prescribed classes of medication worldwide.

These clinical effects, proportionate to patients’ risk profile [7], have been credited principally to total-cholesterol- and LDL-cholesterol-lowering [8]; however, statins have been shown also to improve concurrently vascular function, inflammation and oxidative stress, actions that should contribute to their overall cardiovascular benefit [9,10]. An established mechanism for these additional ‘pleiotropic’ actions is via the inhibition of isoprenoid intermediates necessary for small G-protein function that also are synthesized through the mevalonate pathway [11]. At the intracellular level, small G-proteins, such as Ras, Rac1 and RhoA, participate in several signalling pathways with cardiovascular implications (see [9]). These pathways are involved in modulating the production of ROS (reactive oxygen species), through NADPH oxidase [12] and ET-1 (endothelin-1) [13,14], and, by destabilizing and uncoupling eNOS (endothelial NOS [nitric oxide synthase]) mRNA, the availability of the antioxidant vasodilator NO (nitric oxide) [15]. They can also regulate AT1 receptor (angiotensin type 1 receptor) gene expression [16]. AT1
receptor stimulation via AngII (angiotensin II), a bioactive peptide up-regulated in hypertension and heart failure, also increases ROS production [17,18].

Statins exert additional effects on NO bioavailability by activating PI3K (phosphoinositide 3-kinase)/Akt signalling, increasing eNOS phosphorylation [19] and by reducing caveolin-1, a structural membrane protein that binds and inhibits NOS function [20]. The changes in the production and availability of ROS and NO observed with statin therapy are thought to be one mechanism responsible for increasing conduit artery endothelium-dependent vasodilation, promoting angiogenesis, inhibiting vascular smooth cell proliferation, and reducing oxidative stress and inflammation [8–10,21].

The possibility that statins also reduce cardiovascular risk by rectifying the disturbed neural control of the heart and circulation present in many disease conditions has received less attention. The purpose of the present review is to focus on the effects of statin therapy on the autonomic nervous system, with particular emphasis on human studies measuring post-ganglionic efferent MSNA (muscle sympathetic nerve activity) and HRV (heart rate variability).

**EXPERIMENTAL BACKGROUND**

In addition to the vasculature, the small G-proteins described above are also present and functionally active in central neural sites involved in the generation of sympathetic and parasympathetic outflow, such as the RVLM (rostral ventrolateral medulla), nucleus tractus solitarius and paraventricular nucleus. In experimental models of hypertension or heart failure, one or more of these central brainstem regions exhibit elevated oxidative stress with increased ROS [e.g. TBARS (thiobarbituric acid-reacting substance) levels and O$_2^-$ (superoxide anion)] [18,22–25], and reduced NO [26,27] and antioxidant defences [22,28], all potential targets of statin therapy. Increased ROS, such as O$_2^-$, inactivates NO, an important signalling molecule in the brain [29], and produce the highly reactive and cytotoxic peroxynitrate [30]. ROS also balance excitatory and inhibitory amino acids in the RVLM [31], facilitating premotor sympathetic neuron input modulation [32]. As one consequence, inhibitory GABA ($\gamma$-aminobutyric acid) is reduced in the RVLM of hypertensive rats [33,34].

One mechanism responsible for such increased central oxidative stress may be the pathological activation of the RAAS (renin–angiotensin–aldosterone system), which up-regulates and acts on central AT$_1$ receptors to increase NADPH oxidase and mitochondrion-produced ROS [18,24,25,35–37]. Pharmacological interventions designed to inhibit the central effects of aldosterone, AngII or AT$_1$ receptor binding each reduce sympathetic outflow [38–43]. Inflammatory cytokines produced by brain perrivascular macrophages may also be involved [44], increasing NADPH oxidase and O$_2^-$, and up-regulating AT$_1$ receptor activity [45].

A causal role for the central neural imbalance between ROS and NO/antioxidants as a mediator of augmented sympathetic outflow and cardiac vagal tone has been tested similarly using an icv (intracerebroventricular) infusion to exclude the involvement of peripheral afferent neural mechanisms. In animal models of hypertension icv infusion of an NADPH oxidase inhibitor or tempol, a SOD (superoxide dismutase) mimetic given to reduce central ROS production, decreases both RSNA (renal sympathetic nerve activity) and BP (blood pressure) [24,34,46]. Reductions in RSNA with icv tempol involve NO-mediated release of GABA within the RVLM [34,47]. Conversely, reducing NO by icv NOS inhibition using L-NMMA ($N^\text{G}$-monomethyl-L-arginine) increases sympathetic outflow [34]. In healthy rats, the increased RSNA and BP elicited by icv L-NMMA is abolished by transecting the spinal cord at C1 to C2, or by intravenous infusion of L-arginine, a precursor to NO [48,49].

Thus, if brain penetration is achieved, statins could exert sympathetic inhibitory actions centrally by reducing the ROS-mediated central oxidative stress and by increasing production or availability of NO [50,51]. If so, these auxiliary properties could have important applications for patients with conditions such as hypertension, post-myocardial infarction, chronic kidney disease, heart failure and diabetes in whom elevated sympathetic activity and decreased vagal HR (heart rate) modulation are present, and are associated with increased risk of ventricular hypertrophy [52], impaired endothelium-dependent and independent vasodilation [53–55], insulin resistance [56,57], arrhythmias [58] and premature mortality [59–61].

Individual statins differ with respect to lipophilicity, which influences their tissue-selectivity. Hydrophilic statins such as rosuvastatin and pravastatin undergo primarily hepatic uptake by a carrier-mediated organic anion transport polypeptide, whereas lipophilic statins (simvastatin, atorvastatin, fluvastatin and lovastatin) are able to diffuse passively across extrahepatic cell membranes [62]. Consequently, lipophilic statins have a greater potential to cross the blood–brain barrier and interact with central brain regions involved in the generation or modulation of efferent sympathetic and parasympathetic activity. To date, because the majority of studies investigating potential autonomic effects have administered lipophilic statins, data concerning hydrophilic statins are limited.

**EXPERIMENTAL STUDIES**

The effects of statin therapy on autonomic nervous system function have been studied thus far in experimental models of human hypercholesterolaemia, diabetes, the metabolic syndrome, hypertension, myocardial infarction and heart failure.

**Hypercholesterolaemia**

In genetically dyslipidaemic apoE (apolipoprotein E)$^{-/-}$ mice, 2 weeks of oral rosuvastatin (80 mg/kg of body weight per day) had no effect on LDL-cholesterol, but the HF (high-frequency) power of HRV, BP and HR reverted to values of control mice [20]. Rosuvastatin also increased the HR response to atropine and the SBP (systolic BP) response to NOS inhibition, suggesting increased arterial baroreflex modulation.
Diabetes
In streptozotocin-induced diabetic rats, 2 weeks of oral high-dose fluvastatin (10 mg/kg of body weight per day) did not alter total cholesterol, but normalized cardiac sympathetic MIBG (methyldobenzylguanidine) scintigraphy [63]. Compared with healthy control animals, MIBG accumulation was reduced in untreated diabetic animals, but normalized with fluvastatin, although the two groups exhibited a similar washout rate, a marker of decreased NE (noradrenaline) turnover and thus cardiac sympathetic activity [64]. The difference in MIBG accumulation may then be related to impaired NE neural uptake. Fluvastatin did not alter BP but reduced markers of oxidative stress, plasma levels of lipid peroxides, and mRNA expression of myocardial 8-isoprostaglandin F2α, and NADPH oxidase subunit p22phox [63].

Metabolic syndrome
Silva et al. [65] reported that 2 weeks of oral simvastatin (5 mg/kg of body weight per day) altered HR responses to methylatropine and propanolol in rats with the metabolic syndrome: the increase in HR following methylatropine was augmented and the decrease in HR following propanolol was diminished, suggesting increased cardiac vagal tone and reduced sympathetic HR modulation. Simvastatin improved insulin resistance but did not change BP.

Hypertension
In SHRSPs (stroke-prone spontaneously hypertensive rats), 30 days of high-dose oral atorvastatin (50 mg/kg of body weight per day) reduced 24-h urinary NE excretion and mean arterial pressure [66]. These effects were not observed in control WKY (Wistar–Kyoto) rats even though atorvastatin reduced total cholesterol and LDL-cholesterol in both groups. Atorvastatin increased the expression of eNOS (cortex, cerebellum, hypothalamus, brainstem and aorta) and inducible NOS (cortex, hypothalamus, brainstem and aorta) in both the SHRSP and WKY groups, but no effect on nNOS (neuronal NOS) was detected. The same effects were not observed in normolipidaemic rabbits with pacing-induced ventricular tachyarrhythmias compared with vehicle-treated infarcted animals [75].

Post-myocardial infarction
In an experimental rat model of myocardial infarction, 4 weeks of pravastatin (5 mg/kg of body weight per day) did not alter total cholesterol or NE (plasma or tissue), but reduced cardiac sympathetic MIBG accumulation, and pacing-induced ventricular tachyarrhythmias compared with vehicle-treated infarcted animals [75].

Heart failure
Following 3 weeks of oral simvastatin at a dose of 1.5 or 3 mg/kg of body weight per day (but not 0.3 mg/kg of body weight per day), Pliquett et al. [76] reported lower RSNA and plasma NE concentrations in normolipidaemic rabbits with pacing-induced heart failure compared with a vehicle-treated group. The arterial baroreflex modulation of RSNA and of HR in response to sodium nitroprusside (but not phenylephrine) infusion were also normalized in the simvastatin-treated group (1.5 mg/kg of body weight per day and 3 mg/kg of body weight per day). Fractional shortening remained depressed. The 3 mg/kg of body weight per day dose restored both time [SDNN (S.D. of consecutive normal R-wave to R-wave intervals)] and frequency (total spectral power) domain measures of HRV in the heart failure group to values obtained in control animals [77].

Briefer durations of statin therapy may also be sufficient to achieve such autonomic effects. In an experiment by Gao et al.
[78] again involving normolipidaemic heart failure rabbits, 7 days of simvastatin (3 mg/kg of body weight per day) resulted in lower RSNA and total peripheral resistance. Mean LVEF (left ventricular ejection fraction) was 78% in non-heart failure controls, 32% in rabbits with heart failure given placebo and 52% in those treated with simvastatin (P < 0.05 compared with placebo). Simvastatin also increased arterial baroreflex gain for both RSNA and HR modulation, and reduced the RSNA and BP responses to acute icv AngII. Resting BP and HR were unchanged. In the animals treated with simvastatin, RVLVM AT1 receptor and NADPH oxidase subunit (p91phox, p47phox, and p40phox) mRNA and protein expression, and NADPH-oxidase-dependent O2* production were reduced. No significant adaptations were found in animals treated with both simvastatin and icv AngII.

In a subsequent study involving normolipidaemic rabbits with heart failure, 7 days of icv simvastatin infusion (5 μg/0.5 μl per h) reduced RSNA and increased the arterial baroreflex gain of RSNA [51]. There was no effect on LVEF. Central NOS inhibition with icv l-NAME prevented these RSNA responses. Simvastatin treatment increased nNOS protein expression in the RVLVM. The authors provided in vitro confirmation that 0.1 and 1 μM, but not 10 nM, simvastatin increased nNOS mRNA expression; this effect was blocked by co-administration of upstream isoprenoid intermediates t-mevalonate, farnesyl pyrophosphate or geranylgeranyl pyrophosphate, and was replicated by administration of a ROCK (Rho-associated kinase) inhibitor. These observations demonstrate that simvastatin inhibits the mevalonate pathway and implicate the RhoA/ROCK pathway in statin-induced up-regulation of nNOS. Recent work with icv fasudil, a ROCKII inhibitor, confirms an NO-dependent improvement in parasympathetic and sympathetic modulation in rabbits with heart failure [79].

In a rat model of chronic heart failure, 15 days of oral fluvastatin (10 mg/kg of body weight per day) increased arterial baroreflex control of HR and cardiac output compared with untreated animals [80]. Although HR and BP were unchanged, fluvastatin decreased cardiac hypertrophy and normalized markers of oxidative stress (decreased serum malondialdehyde and reduced glutathione; increased SOD and inflammation (decreased TNF-α (tumour necrosis factor-α)).

Collectively, these results suggest that high-dose lipophilic statins can alter autonomic function in a variety of experimental models of human disease. These effects appear to be mediated by improvements in central oxidative stress through their actions on ROS production and antioxidant defences. The increased sensitivities of arterial baroreflex regulation of RSNA and HR described by several investigators [51,68,76,78] could also implicate an afferent peripheral statin mechanism, for example via increased vascular NO bioavailability improving conduit artery compliance and baroreceptor signal transduction. However, overexpression of eNOS or nNOS by gene transfer into the RVLVM also improves the baroreflex control of HR and RSNA in hypertensive and heart failure animals [81,82], and, since augmentation of the reflex control of RSNA was effected by icv infusion of simvastatin [51], a central neural reduction in oxidative stress may be sufficient to account for this finding. With evidence that low-dose simvastatin (0.25 mg/kg of body weight) can improve the 28-day survival of mice subjected to myocardial infarction [83], further work should be directed to establishing whether autonomic effects contribute to this benefit.

### HUMAN STUDIES

The autonomic effects of statin therapy have been investigated in patients with hypercholesterolaemia, hypertension, coronary artery disease, chronic kidney disease and heart failure. These data are summarized in Tables 1 and 2.
### Table 2 Effects of statin therapy on HRV in humans

<table>
<thead>
<tr>
<th>Trial reference</th>
<th>Population</th>
<th>Study design</th>
<th>Subjects (n)</th>
<th>Intervention (dose; duration)</th>
<th>Major HRV findings</th>
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<tbody>
<tr>
<td>[84]</td>
<td>HC</td>
<td>RC</td>
<td>29</td>
<td>Atorvastatin (10 mg/day; 10 weeks)</td>
<td>↑SDNN, RMSSD, TP and VLF</td>
</tr>
<tr>
<td>[88]</td>
<td>HC</td>
<td>UC</td>
<td>74</td>
<td>Atorvastatin (40 mg/day; 56 weeks)</td>
<td>No change in HRV</td>
</tr>
<tr>
<td>[89]</td>
<td>HC</td>
<td>R</td>
<td>31</td>
<td>Simvastatin (20 mg/day; 6 weeks)</td>
<td>No change in HRV</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>30</td>
<td>Simvastatin + omega-3</td>
<td>No change in HRV</td>
</tr>
<tr>
<td>[90]</td>
<td>HC</td>
<td>RC</td>
<td>26</td>
<td>Pravastatin (20 mg/day; 8 weeks)</td>
<td>↑Night time HF power</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Simvastatin (20 mg/day; 8 weeks)</td>
<td>No change in HRV</td>
</tr>
<tr>
<td>[93]</td>
<td>HTN</td>
<td>RPCC</td>
<td>13</td>
<td>Atorvastatin (80 mg/day; 3 weeks)</td>
<td>No change in HRV</td>
</tr>
<tr>
<td>[94]</td>
<td>HTN + SN</td>
<td>RPCC</td>
<td>14</td>
<td>Simvastatin (80 mg/day; 4 weeks)</td>
<td>No change in HRV</td>
</tr>
<tr>
<td>[95]</td>
<td>HC ± CAD</td>
<td>CON</td>
<td>S: 40</td>
<td>Atorvastatin (20 mg/day; 112 weeks)</td>
<td>↑SDNN, pNN50, RMSSD and TP and ↓LF and LF/HF ratio</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td>C: 20</td>
<td>None</td>
<td></td>
</tr>
<tr>
<td>[96]</td>
<td>CAD</td>
<td>RPCC</td>
<td>10</td>
<td>Atorvastatin (80 mg/day; 4 weeks)</td>
<td>Trend for ↓LF/HF ratio</td>
</tr>
<tr>
<td>[97]</td>
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<td>OC</td>
<td>S: 29</td>
<td>Any statin</td>
<td>↑SDNN</td>
</tr>
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<td></td>
<td></td>
<td>C: 44</td>
<td>None</td>
<td></td>
</tr>
<tr>
<td>[98]</td>
<td>CAD</td>
<td>RPCC</td>
<td>80</td>
<td>Simvastatin (80 mg/day; 6 weeks)</td>
<td>No change in HRV</td>
</tr>
<tr>
<td>[99]</td>
<td>CAD</td>
<td>OC</td>
<td>S: 54</td>
<td>Any statin</td>
<td>No change in HRV</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td>C: 32</td>
<td>None</td>
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<tr>
<td>[102]</td>
<td>CKD</td>
<td>RPCC</td>
<td>10</td>
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</tr>
<tr>
<td>[106]</td>
<td>CHF</td>
<td>RCT</td>
<td>S: 40</td>
<td>Atorvastatin (10 mg/day; 12 weeks)</td>
<td>↑SDNN and RMSSD</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td>C: 40</td>
<td>None</td>
<td></td>
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<tr>
<td>[107]</td>
<td>CHF</td>
<td>RCT</td>
<td>S: 12</td>
<td>Atorvastatin (40 mg/day; 12 weeks)</td>
<td>↓LF and LF/HF ratio</td>
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<td></td>
<td>C: 9</td>
<td>Placebo</td>
<td></td>
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<tr>
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<td>CHF</td>
<td>RPCC</td>
<td>15</td>
<td>Atorvastatin (80 mg/day; 12 weeks)</td>
<td>No change in HRV</td>
</tr>
<tr>
<td>[112]</td>
<td>CHF</td>
<td>UC</td>
<td>25</td>
<td>Simvastatin (20 mg/day; 6 weeks)</td>
<td>No change in HRV</td>
</tr>
</tbody>
</table>

### Hypercholesterolaemia

In a randomized cross-over trial of 29 patients with hypercholesterolaemia, Melenovsky et al. [84] compared the effects of 10 weeks of low-dose atorvastatin (10 mg/day) or fenofibrate (200 mg/day) on HRV. Both atorvastatin and fenofibrate reduced total cholesterol and increased time domain [SDNN and RMSSD (square root of the mean square difference of successive RR intervals)] and frequency domain (total spectral power and very-LF power) measures of HRV. However, interpretation of these findings is limited by the lack of a placebo control group. Atorvastatin also increased the arterial baroreflex sensitivity of HR, as did 6 weeks of diet plus atorvastatin (10 mg/day) in a small cross-over trial of ten otherwise healthy men with elevated total cholesterol [85]. In a controlled study of 30 hypercholesterolaemic patients with Type 2 diabetes, 12 months of simvastatin (40 mg/day) reduced total cholesterol and LDL-cholesterol, and increased HR recovery, a marker of parasympathetic potency [86], 1 min following a maximal exercise test [87].

In contrast, in an uncontrolled trial of 74 hypercholesterolaemic men, 12 months of atorvastatin (40 mg/day) reduced total cholesterol and LDL-cholesterol, but did not affect time or frequency domain measures of HRV [88]. Similar neutral results have been observed following 6 weeks of simvastatin (20 mg/day) or simvastatin (20 mg/day) plus omega-3 fatty acid supplementation [89]. However, both studies are again confounded by the absence of a control group.

Interestingly, in a randomized cross-over trial of 26 hyperlipidaemic patients, night-time HF power, a marker of parasympathetic modulation, was increased following 8 weeks of pravastatin (20 mg/day), but not simvastatin (20 mg/day) [90], suggesting a sinoatrial action of this more hydrophilic statin at a time of low cardiac sympathetic tone.

### Hypertension

The most consistent evidence in humans for a sympathoinhibitory role of statins arises from patients with primary hypertension. Sinski et al. [91] studied ten hypercholesterolaemic essential hypertensive men and found that, compared with an untreated control group, 8 weeks of atorvastatin (20 mg/day) reduced MSNA burst incidence and frequency by 17% (46.5 ± 6.2 to 38.4 ± 5.5 bursts/100 heart beats; 36.0 ± 6.6 to 28.6 ± 4.8 bursts/min), without altering BP or heart rate. Subsequently this group conducted a larger randomized double-blind placebo-controlled study of simvastatin (40 mg/day for 8 weeks) in 31 hypercholesterolaemic hypertensive men [92]. Those receiving simvastatin (n = 15) exhibited significant reductions in MSNA burst...
incidence (18%: 47.8 ± 8.4 to 39.4 ± 8.3 bursts/100 heart beats) and burst frequency (24%: 36.5 ± 5.4 to 27.8 ± 5.8 bursts/min), and HR (8%: 77 ± 7 to 71 ± 5 beats/min). BP was unchanged. None of these variables changed significantly in the placebo group. In both studies the statin treatment was reported to increase arterial baroreflex sensitivity of HR, a marker of reflex cardiac vagal modulation [91,92].

Using a more stringent randomized double-blind placebo-controlled cross-over study design, Gomes et al. [93] reported significant mean 10% and 11% reductions in MSNA burst incidence (64.7 ± 3.0 to 58.5 ± 2.0 bursts/100 heart beats) and burst frequency (39.2 ± 1.5 to 35.0 ± 2.0 bursts/min) following 3 weeks of atorvastatin (80 mg/day) in 13 primary hypertensive patients. Neither 24-h ambulatory BP nor resting HR or HRV was affected. Of note, because four out of their 13 patients had discontinued pre-existing therapy before their participation, these values may have been affected by a residual central neural statin action.

To avoid potential confounding effects of hypercholesterolaemia or previous statin therapy we conducted a randomized double-blind placebo-controlled cross-over study investigating the effects of 4 weeks of simvastatin (80 mg/day) on MSNA in 14 essential hypertensive normolipidaemic statin-naïve patients who otherwise would not have received statin therapy [94]. Simvastatin lowered mean MSNA burst incidence (55 ± 23 to 43 ± 17 bursts/100 heart beats) and burst frequency (32 ± 12 to 25 ± 9 bursts/min) by 22%. Representative tracings from two patients are demonstrated in Figure 1. Simvastatin did not alter HRV or arterial baroreflex sensitivity of HR and MSNA, ruling out an effect on efferent sinoatrial node discharge or afferent peripheral mechanisms of action. These results are in agreement with experimental findings suggesting a central autonomic action of statins. In three patients, simvastatin reduced atrial or ventricular ectopy by 55%. There were no significant effects on the homoeostatic model assessment of insulin resistance, BP or HR. Interestingly, with simvastatin a trend was observed for a reduction in DBP (diastolic BP) (81 ± 10 to 77 ± 8; P = 0.055), a response which should normally trigger reflexive increases in MSNA. Simvastatin increased also endothelium-independent vasodilation, possibly as a result of less neurogenic constraint, but did not alter endothelium-dependent (flow-mediated) vasodilation.

**Coronary artery disease**

In a controlled trial of 40 hypercholesterolaemic patients with and without coronary artery disease, 24 months of atorvastatin (20 mg/day) reduced total cholesterol and LDL-cholesterol, and altered time domain [increased SDNN, RMSSD and pNN50 (percentage of consecutive normal RR intervals that differ by more than 50 ms)] and frequency domain (increased total spectral power and HF power; decreased LF power and the LF/HF ratio).
ratio) measures of HRV [95]. Atorvastatin did not change HR or BP.

In a smaller randomized double-blind placebo-controlled cross-over study of ten patients with documented coronary artery disease, 4 weeks of atorvastatin (80 mg/day) reduced resting plasma NE and demonstrated a trend towards a reduced LF/HF ratio of HRV [96]. These authors reported no significant change in plasma NE or HRV responses to supine tilt (80°) or in pressor or HR responses to a cold pressor test. Unfortunately, these subjects were not statin-naïve but underwent a 4-week medication washout period prior to participation. Increased HRV has also been reported in a controlled observational study of 29 post-infarct patients on statins [97].

In contrast, in a larger randomized double-blind placebo-controlled cross-over trial of 80 patients with diagnosed coronary artery disease, 6 weeks of high-dose atorvastatin (80 mg/day) reduced total cholesterol and LDL-cholesterol, but did not alter time domain HRV [98]. Similar neutral HRV results have been observed in a controlled observational study of 54 coronary artery disease patients with an implanted cardioverter defibrillator on statins [99].

**Chronic kidney disease**

In a randomized-controlled trial of ten hypertensive patients with chronic kidney disease, Siddiqui et al. [100] observed that, compared with usual-care controls, 6 weeks of atorvastatin (20 mg/day) reduced MSNA burst incidence by 30% (50 ± 13 to 35 ± 12 bursts/100 heart beats) and burst frequency by 29% (28 ± 8 to 20 ± 6 bursts/min). Atorvastatin did not lower LDL-cholesterol or total cholesterol, BP or HR. It is conceivable that greater MSNA reductions might have been detected had these patients not been pre-treated for hypertension with aliskiren, a renin inhibitor known to reduce MSNA in this population [101].

In contrast, in a randomized double-blind placebo-controlled cross-over study, 4 weeks of simvastatin (40 mg/day) lowered LDL-cholesterol, but did not affect time domain measures of HRV in ten chronic haemodialysis patients [102].

**Chronic heart failure**

In comparison with the consistent reductions in RSNA in experimental preparations [51,76,78], thus far studies in human heart failure have yielded less conclusive results. Gomes et al. [103] prospectively examined eight heart failure patients [NYHA (New York Heart Association) classes II–IV and LVEF <40%] before and 8 weeks after discontinuation of atorvastatin or simvastatin (40 mg/day). They reported a significant increase in mean MSNA burst incidence (56 ± 5 to 73 ± 4 bursts/100 heart beats) and frequency (32 ± 3 to 42 ± 3 bursts/min). Values returned to baseline levels 4 weeks after statins were re-prescribed (burst incidence, 52 ± 6 bursts/100 heart beats; frequency, 28 ± 3 bursts/min).

In a double-blind randomized-controlled trial of 18 statin-naïve non-ischaemic systolic heart failure patients (LVEF <35%), Horwich et al. [104] studied the effects of 12 weeks of low-dose atorvastatin (10 mg/day) (n = 9) or placebo (n = 9) on MSNA. Atorvastatin lowered both LDL-cholesterol and total cholesterol, but did not produce a statistically significant reduction in MSNA, although burst incidence (62 ± 6 to 52 ± 6 bursts/100 heart beats) and frequency (43 ± 3 to 36 ± 5 bursts/min) fell by 16% with statin therapy compared with ~3% reductions in the placebo group. Inspection of the individual data demonstrates that this mean MSNA change was driven primarily by one subject with a very large reduction (>30 bursts/min). Discrepancy in these published findings suggest that the autonomic effects of statins are not universal in heart failure, but are unrelated to clear-cut cholesterol reductions. They may also raise important questions regarding a dose threshold for the autonomic effects of statins in a heart failure population.

More recently, Deo et al. [105] reported the findings of a series of investigations in patients with idiopathic dilated cardiomyopathy (NYHA classes I–III and LVEF <40%). In both groups, simvastatin reduced both LDL-cholesterol and total cholesterol. In the initial proof-of-concept study involving seven patients, 4 weeks of simvastatin (40 mg/day) reduced mean MSNA burst incidence and frequency by 12–13% (75 ± 5 to 65 ± 5 bursts/100 heart beats; 50 ± 4 to 44 ± 5 bursts/min), and also MAP. In a subsequent double-blind placebo-controlled cross-over study of six patients, simvastatin reduced mean MSNA burst incidence (24%; 59 ± 5 bursts/100 heart beats in placebo to 45 ± 6 bursts/100 heart beats in statin-treated) and frequency (27%; 44 ± 6 bursts/min in placebo to 32 ± 5 bursts/min in statin-treated), and also total ROS and O2•−, as estimated from plasma by electron paramagnetic spin resonance. These findings are consistent with the concept that reductions in sympathetic activity with lipophilic statins occur secondarily to a reduction in oxidative stress.

In a randomized-controlled trial, Vrtovjek et al. [106] reported that heart failure patients (LVEF <30%) receiving 3 months of atorvastatin (10 mg/day; n = 40) had increased time domain HRV (SDNN and RMSSD) and QT interval variability compared with controls. They reported that these effects were independent of the reductions in total cholesterol. In a single-blind randomized-controlled trial of 21 systolic heart failure patients (LVEF <45%), Hamaad et al. [107] reported that 12 weeks of atorvastatin (40 mg/day) reduced the LF power of HRV, and consequently the LF/HF ratio. The significance of this finding is unknown in so far as there is an inverse relationship between MSNA and LF spectral power as heart failure progresses [108], and in a multivariate analysis involving heart failure patients reduced daytime LF power of HR was an independent predictor of increased risk of sudden cardiac death [109]. In a controlled study, 3 months of fluvastatin (80 mg/day) reduced total cholesterol and LDL-cholesterol, and increased HR recovery at 1 and 3 min following a maximal exercise test in 29 hyperlipidaemic patients with ischaemic cardiomyopathy (LVEF <40%) [110].

In contrast, in a randomized double-blind placebo-controlled cross-over study of 15 non-ischaemic cardiomyopathy patients (LVEF <40%), 12 weeks of atorvastatin (80 mg/day) reduced LDL-cholesterol, but did not alter HRV [111]. Similar neutral HRV results have been reported in an uncontrolled trial of 25 patients with non-ischaemic dilated cardiomyopathy (LVEF <40%) following 6 weeks of simvastatin (20 mg/day) [112]. However, this group did not report an intriguing correlation, with an inverse
relationship between the changes in baseline LDL-cholesterol following simvastatin therapy and the changes in LF power during a Valsalva stress.

In the only head-to-head trial of a lipophilic statin, Tsutamoto et al. [113] compared 6 months of low-dose atorvastatin (5 mg/day) or rosuvastatin (2.5 mg/day) on several end points in non-diabetic patients with dilated cardiomyopathy (LVEF <45%). Total cholesterol and LDL-cholesterol were reduced equally in both groups. Although plasma NE was unchanged, in the atorvastatin group cardiac sympathetic imaging using MIBG scintigraphy detected an increased H/M (heart/mediastinum) ratio (a marker of increased NE uptake or cardiac adrenoreceptor density [114]) and a decreased washout rate. Additionally, the atorvastatin group reported increased LVEF and reduced plasma NT-proBNP (N-terminal pro-B-type natriuretic peptide) and oxidized LDL-cholesterol concentrations. A trend for reduced hsCRP (high-sensitivity C-reactive protein) was also observed.

DISCUSSION AND IMPLICATIONS

The available human evidence indicates that several statins are capable of exerting autonomic effects. The most consistent finding is that of a significant reduction in efferent sympathetic outflow, measured as MSNA, with lipophilic statins in subjects with normal ventricular function. Simvastatin and atorvastatin, across a wide range of doses, reduced MSNA by 10–24% in patients with hypertension (Table 1). Low-dose atorvastatin failed to reduce MSNA in one study of heart failure patients [104], although overall, when studied in heart failure, MSNA fell by 12–29%.

In contrast with these generally concordant findings, only ~50% of the studies published reported a significant change in HRV with statin therapy. This lack of consensus probably results from differences between investigative laboratories with respect to the time and frequency domain methods they use to infer vagal and sympathetic HR modulation; differences in the quality of study designs (Table 2); the sample size implication of the skewed (non-parametric) distribution of such variables within groups (i.e. some studies may have lacked the statistical power to demonstrate true differences) [115]; the impact of heart failure on the congruence of MSNA and LF spectral power [108,116]; the confounding potential of background medication with effect on sinoatrial discharge rates, such as \(\beta\)-adrenergic antagonists [116–118]; and differences in statin dosage. For example, a prospective trial involving 359 subjects reported significant positive correlations between statin dose and the time domain indices of vagal HR modulation (SDNN and RMSSD) studied [119].

Although limited mechanistic human data are available, based on experimental evidence, the autonomic effects appear to be mediated by a reduction in central oxidative stress, secondary to changes in NO and ROS production and availability. Penetration of statins into brain regions associated with generating and modulating sympathetic and parasympathetic outflow may be dependent on statin lipophilicity. However, the effects of hydrophilic statins on MSNA or HRV have not been thoroughly tested.

The published literature raises several important questions as to the clinical implications of the autonomic changes effected by statin therapy.

Is there a relationship between MSNA and BP or insulin resistance?

Notable in these small clinical trials demonstrating a fall in MSNA has been the absence of reductions in resting or ambulatory BP. This has raised the question as to whether this effect of lipophilic statins on MSNA is clinically relevant [120]. In a randomized double-blind placebo-controlled trial of 973 healthy normotensive subjects, 6 months of simvastatin (20 mg/day) or pravastatin (40 mg/day) both reduced SBP and DBP equally (decrease of 2.4–2.8 mmHg) [121]. Meta-analyses have confirmed long-term statin usage can produce modest reductions in resting BP (decrease by ~2–4/1–2 mmHg), with the largest effects observed in patients with elevated BP [122,123]. These reductions also appear to be independent of changes in plasma lipids [124]. Current studies of autonomic effects of statins have been primarily of small cohorts of patients receiving anti-hypertensive medications or with normal BP, i.e. groups unlikely to demonstrate substantive reductions in BP. Future investigations of this question would benefit from longer treatment periods and adequate statistical power to detect modest BP changes.

A relationship between sympathetic activity and insulin resistance has been demonstrated in a number of well-controlled interventional studies. Insulin resistance is reduced with acute increases in MSNA in healthy normotensive men [125] and improved following treatment with \(\alpha\)-adrenergic blockers [126,127] or central-acting sympatholytics in heart failure or hypertensive populations [128–130]. Indeed, positive correlations have been reported between MSNA and insulin resistance in obese patients [57]. Given that statins can be a robust stimulus to lower MSNA [129], this could mean that patients with low baseline sympathetic activity, and thus presumably less likely to experience large reductions in MSNA, are more susceptible to a statin-mediated increase in insulin resistance. Interestingly, in the only study to simultaneously examine MSNA and insulin sensitivity with statin therapy, McGowan et al. [94] found no change in insulin resistance after 4 weeks of simvastatin in normolipidaemic primary hypertensive patients, even though MSNA was reduced. The interactive autonomic and metabolic consequences of statin therapy requires further study.

Are the mechanisms responsible for the autonomic effects cholesterol-independent?

The experimental evidence suggests that statins exert autonomic effects via non-cholesterol-mediated or ‘pleiotropic’ actions on
central oxidative stress. However, not all human studies have been designed specifically to test whether MSNA reductions are indeed cholesterol-independent. Although the majority of studies (Table 1) have observed parallel reductions in LDL-cholesterol and MSNA, one study reported reductions in LDL-cholesterol without corresponding reductions in MSNA [104], while another reported significant reductions in MSNA, but only a trend towards cholesterol-lowering [100]. Correlation analyses have also failed to detect a significant association between the changes in LDL-cholesterol or total cholesterol and MSNA, but the individual study sample sizes are small.

Acute lipid infusion can increase MSNA in both young and old participants [132,133]. One group has reported a positive significant correlation between total cholesterol and MSNA burst incidence ($r = 0.50$) [93]. Treatment with a $\alpha_1$-adrenoreceptor inhibitor has been shown to lower total cholesterol and LDL-cholesterol in hypertensive patients [134], whereas 4 weeks of post-ganglionic sympathetic neuron blockade with debrisoquine caused significant, correlated, changes in plasma NE and total cholesterol in essential hypertensive patients, but not healthy controls [135]. McGowan et al. [94] found the change in MSNA elicited by simvastatin to correlate with pre-treatment total cholesterol and LDL-cholesterol ($r = 0.56$ and 0.59; i.e. the higher baseline total cholesterol or LDL-cholesterol the greater the change in MSNA). However, whether this relationship was causal or coincidental could not be determined definitively. Total cholesterol and LDL-cholesterol have been found to associate negatively with HRV [136], whereas the sympathetically mediated morning surge in BP [137] correlates positively with plasma LDL-cholesterol concentrations [138]. Further work is required to confirm the cholesterol-independence of the observed statin autonomic effects.

**Do these actions influence cardiovascular end points?**

Both increased MSNA and decreased HRV have been linked to adverse cardiovascular events [59–61]. Thus impaired neurogenic circulatory regulation may be an additional mechanism by which statins reduce cardiovascular morbidity and mortality. However, several large-scale trials [e.g. GISSI-HF (Gruppo Italiano per lo Studio della Streptochinasi nell’Infarto Miocardico-Heart Failure), CORONA (Controlled Rosuvastatin Multinational Study in Heart Failure) and AURORA (A Study to Evaluate the Use of Rosuvastatin in Subjects On Regular Haemodialysis: an Assessment of Survival and Cardiovascular Events) in heart failure or chronic kidney disease populations have reported that statins do not improve cardiovascular events despite lowering LDL-cholesterol and total cholesterol [139–141]. Interestingly, in each of these large neutral trials the statin tested was rosuvastatin, a hydrophilic statin. Lipophilicity may be the key to these neutral outcomes. In a study involving patients with dilated cardiomyopathy, 6 months of atorvastatin, but not rosuvastatin, improved cardiac MIBG scintigraphy parameters, LVEF, NT-proBNP and oxidized LDL-cholesterol concentrations [113]. In contrast, in a recently published 4-year follow-up of Japanese patients with chronic heart failure, pitavastatin, a lipophilic statin, at 2 mg/day (equivalent to simvastatin at approximately 20 mg/day or atorvastatin at approximately 10 mg/day) did not improve cardiac mortality and hospitalization for worsening heart failure [142]. Predefined subgroup analysis did demonstrate improvements in patients with a LVEF >30%, suggesting a possible statin–ventricular function interaction. A limitation of that study is that, although the dose was sufficient to reduce LDL-cholesterol, it may not have been high enough to permit adequate penetration of central neural sites and produce autonomic effects. Indeed, the only human trial not to demonstrate a statistically significant reduction in MSNA used a similar dose of atorvastatin (10 mg/day) [104]. The concept that higher doses of lipophilic agents may be required to act on central neural sites has been put forth for AT1 receptor blockers [143]. The issue of lipophilicity and statin penetration is complicated by evidence that hydrophilic statins are capable of reducing circulating isoprenoid levels as a result of hepatic HMG-CoA reductase inhibition [144]. Future work is required to elucidate possible differences in clinical benefits of lipophilic and hydrophilic statins.

Alternatively, the neutral finding of lipophilic statin therapy in human heart failure may represent the net of concurrent positive and deleterious actions. Evidence for an adverse role of low cholesterol has been documented in heart failure, with mortality increasing 25% for each mmol/l decrease in total cholesterol [145,146]. Lipophilic statin therapy may also attenuate adaptations in cardiorespiratory fitness following aerobic exercise training [147], a significant predictor of mortality [148,149]. Thus, in some patient populations, the overall statin effect may be the balance of its beneficial and adverse properties.

One likely mechanism by which statin-mediated autonomic effects may improve cardiovascular outcomes is by reducing the risk of arrhythmias and sudden cardiac death. Sympathetic activity has been identified as an important trigger to increase the dispersion of repolarization or the generation of afterdepolarizations [58]. Likewise reduced autonomic tone, as assessed by decreased HRV, is associated with ventricular arrhythmias and sudden cardiac death [60,150,151]. Statin therapy has been associated with reduced sudden cardiac death in a number of prospective trials [LIPID (Long-term Intervention with Pravastatin in Ischaemic Disease) and 4S (Scandinavian Simvastatin Survival Study), while meta-analyses (>110 000 patients) report that statin therapy decreased the risk of sudden cardiac death by 10% and non-sudden cardiac deaths by 20% [152,153], but did not affect the risk of ventricular tachyarrhythmias or cardiac arrest [154]. These improvements also appear to occur independently of the reductions in LDL-cholesterol.

In contrast, other analyses have reported significant reductions in ventricular tachyarrhythmias with statin therapy [155,156]. Lipophilic, but not hydrophilic, statin therapy can also produce a dose-independent reduction in the incidence and risk of reoccurrence for atrial fibrillation [157]. Statin lipophilicity may explain the discrepancy in some meta-analytic results [158].

Another potential benefit of attenuating central sympathetic outflow in both heart failure and hypertension is reducing renal sodium retention [159]. If this were the case, it might account for reductions in post-acute coronary syndrome hospitalizations for heart failure with atorvastatin reported in the PROVE IT-TIMI 22 (Pravastatin or Atorvastatin Evaluation and Infection Trial-Thrombolysis in Myocardial Infarction 22) trial [160].
CONCLUSIONS

Conventionally, the benefits of statin therapy have been ascribed principally to their effects on plasma cholesterol. However, in conditions characterized by exaggerated sympathoexcitation and elevated oxidative stress, lipophilic statin therapies have demonstrated a consistent capacity to reduce direct effluent sympathetic outflow in both animal and human studies. In experimental preparations, these reductions appear to be mediated by quenching oxidative stress in central brain regions involved in generating or modulating autonomic activity, as a result of reduced ROS and increased NO production. In humans, resting levels of efferent sympathetic activity are associated with mortality in some populations, but the clinical significance of the observed reductions in MSNA with statins remain largely unknown, as they, for example, have not been accompanied by parallel decreases in BP or HR. Future research should be directed at determining mechanisms responsible for statin-mediated sympathoinhibition in humans and to establish its potential clinical impact.

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