Neurohormones in an ovine model of compensated postinfarction left ventricular dysfunction

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Rademaker, Miriam T., Vicky A. Cameron, Christopher J. Charles, Eric A. Espiner, M. Gary Nicholls, Christopher J. Pemberton, and A. Mark Richards. Neurohormones in an ovine model of compensated postinfarction left ventricular dysfunction. Am. J. Physiol. Heart Circ. Physiol. 278: H731–H740, 2000.—Clinical heart failure, of, and experimental therapies in, early left ventricular dysfunction that commonly precedes frank heart failure (35). Improved understanding of the pathophysiology of this prefailure phase of left ventricular impairment (and testing of therapeutic interventions) requires an experimental model that reflects the underlying pathology as well as the hemodynamic and neurohormonal profile of the human disease. Clinical heart failure in the developed world (and increasingly in developing countries) is often the result of ventricular dysfunction secondary to myocardial infarction. A model of anterior left ventricular infarction and left ventricular impairment after coronary artery ligation in sheep has previously been reported (22, 23). However, the neurohumoral characteristics of this model have not been described.

Cardiac failure is characterized by the activation of a number of neuroendocrine systems. Some of these are likely to be protective, especially in the short term, whereas deleterious consequences may result from sustained activation of other systems. Advances to date in the prevention and treatment of heart failure have centered on the understanding and manipulation of these underlying hormonal events as exemplified by the development of agents that inhibit the formation or action of angiotensin II (35). In recent years much attention has focused on the natriuretic peptides, circulating hormones primarily of cardiac origin that are significantly elevated after myocardial infarction (11, 33) and in heart failure (25) in proportion to the magnitude of hemodynamic compromise, which show promise as prognostic indicators of these conditions. However, clinical studies investigating myocardial infarction are somewhat limited in that observation cannot begin until the patient is admitted to the hospital, which often occurs many hours or even days after onset of symptoms. In addition, the scope of hormone and hemodynamic measurements is restricted and there is an ethical obligation to provide pharmacological treatment to these patients, which can induce major alterations in both the endocrine and hemodynamic profile. Thus the responses of the natriuretic peptides and other neurohormonal factors both acutely and chroni-
cally after myocardial infarction with compensated cardiac injury remain uncertain.

This study investigates the sequential changes in (and interrelationships between) hemodynamic status and circulating levels of atrial and brain natriuretic peptide (ANP and BNP, respectively) and, for the first time, amino-terminal proBNP (NT-BNP), together with plasma catecholamines, endothelin-1, and the renin-angiotensin-aldosterone system after acute myocardial infarction and progression to compensated cardiac dysfunction in sheep.

**METHODS**

This study was approved by the Animal Ethics Committee of the Christchurch School of Medicine. Twelve Coopworth ewes [body weight: control (n = 4) 44.6 ± 1.6 kg, ligated (n = 8) 43.0 ± 1.2 kg] underwent cannulation of the left carotid artery and jugular vein 2 days before coronary artery ligation. The sheep were anesthetized initially with diazepam (1 mg/kg) and ketamine (4 mg/kg). A 7-Fr sheath with a side-port extension (Cordis, Miami, FL) was inserted into the left carotid artery for subsequent measurement of arterial pressure and baseline blood sampling. A preligation left ventricular angiogram was performed at this time with 20 ml of a mixture of halothane, nitrous oxide, and oxygen; a 7-Fr pigtail catheter introduced into the carotid sheath. The angiograms were recorded on videotape and analyzed on a Siemens ANCOR cine work station (Solna, Sweden) to calculate left ventricular volumes, ejection fraction, and regional wall motion. After ventriculography, the sheep were ventilated on a mixture of halothane, nitrous oxide, and oxygen; a 7-Fr Swan-Ganz thermodilution catheter was placed in the pulmonary artery via the jugular vein for measurement of mean pulmonary arterial and right atrial pressures and cardiac output.

Further instrumentation of the sheep (coronary sinus and left atrial catheterization) and ligation of the coronary arteries were performed via a left lateral thoracotomy as previously described (10). These procedures were carried out under general anesthesia induced by thiopentone sodium (0.7 mg/kg) and maintained with halothane and nitrous oxide, with continuous monitoring of electrocardiograph and arterial pressure. Before ligation a polyvinyl chloride catheter was inserted into the coronary sinus via the left azygous vein for blood sampling; two additional catheters were placed in the left atrium for blood sampling and pressure determination. Immediately before ligation of the coronary arteries, a pack was placed beneath the heart to bring it forward and the coronary circulation was exposed. In 8 of 12 sheep, the homonymous artery (corresponding to the left anterior descending coronary artery in human) and its second diagonal branch were ligated at a point ~40% of the distance from the apex to the base of the heart (22). The remaining four animals did not undergo coronary ligation and served as sham-ligation controls. All 12 sheep received antiarrhythmic medication consisting of 100 mg lidocaine given as a slow intravenous bolus as the pericardium was opened, and an additional 50 mg were given 20 min after the coronary arteries were ligated or at a matched time during sham-ligation procedures (23). Four of eight sheep with ligated coronaries (and 2 controls) also received 1–2 mg atenolol because of ventricular ectopy, and a single animal required direct-current cardioversion for ventricular fibrillation. Perioperative maintenance fluids consisted of 1 liter of 0.9% saline solution delivered over a 2-h period. Postoperative analgesia was provided by 50 mg pethidine given intramuscularly. All catheters remained in place during the 5-wk study. During the study the animals were held in metabolic cages, had free access to water, and ate a mixed diet of chaff and sheep pellets (containing approximately 80 mmol/day sodium, 200 mmol/day potassium).

Hemodynamic measurements (cardiac output, heart rate, mean arterial, pulmonary arterial, and right atrial pressures, total peripheral resistance (mean arterial pressure – right atrial pressure/cardiac output)) were performed 24 and 2 h before coronary artery ligation (baseline) with the animals in a conscious, unpaired state. Ligation of the coronary arteries was then performed (as described above) and, after recovery from anesthesia, additional hemodynamic measurements were made at hours 2, 3, 4, 5, 6, and 12 and days 1, 2, 3, 5, 7, 14, 21, 28, and 35 postligation. Left atrial pressure was sampled from 2 h to 35 days postligation. Arterial, pulmonary, and atrial pressures were measured using Statham pressure transducers (Spectramed Medical Products, Singapore) and an Astromed chart recorder (Astromed, W. Warwick, RI). Heart rate was calculated from the arterial pressure trace.

Cardiac output was measured by thermodilution using the Swan-Ganz catheter connected to a cardiac output computer (Mansfield Scientific, Mansfield, MA). Ten-millilitre aliquots of ice-cold 0.9% saline were delivered down the proximal port of the catheter, and the mean of three values with less than 10% variation were averaged. Arterial blood samples were taken from the carotid sheath 24 and 2 h before coronary artery ligation with the animals in a conscious, unpaired state. The animals were then anesthetized and surgically prepared (as described above) and further samples taken at 30 min and immediately before coronary artery ligation from the left atrial catheter. After ligation, additional blood samples were taken from the left atrium at 30 and 60 min postligation (with the animals still anesthetized) and on recovery from anesthesia, at hours 2, 3, 4, 5, 6, and 12 and days 1, 2, 3, 5, 7, 14, 21, 28, and 35 postligation. Samples were assayed for ANP, BNP, NT-BNP, plasma renin activity, aldosterone, nor-epinephrine, epinephrine (7, 10, 12, 30, 31), and endothelin-1 (see below). Samples for creatine kinase and troponin T were taken up to day 7 postligation and measured with commercially available kits (Boehringer Mannheim, Germany) (26, 38). Additional blood samples were taken from the coronary sinus (control n = 3, ligated n = 5) to give an indication of cardiac peptide production, starting 30 min preligation and ceasing after 7 days postligation because of problems with catheter patency thereafter. These samples were measured for ANP, BNP, NT-BNP, and endothelin. All blood was drawn into tubes on ice, centrifuged at 4°C, and stored at −80°C until analyzed. All samples from each animal were assayed together to avoid interassay variability.

Plasma endothelin-1 levels were measured by radioimmunoassay after extraction based on methods described for ANP (10), except that samples were reincubated for 3 h at room temperature after addition of antisera. Plasma samples (2 ml) were extracted on SepPak C18 cartridges as previously described (14). The extracts underwent radioimmunoassay with a commercially available antisera raised to human endothelin-1 and using human endothelin-1 as a standard (Peninsula Laboratories). Dilutions of the extracts were parallel to human standards, suggesting similar immunological identity. Detection limit of the assay was 1.1 pmol/l. Intra- and interassay coefficients of variation (CVs) were 5.9 and 8.7%, respectively. Recovery of human endothelin-1 spiked into sheep plasma was 69%.

Thirty-five days postligation, an additional left ventricular angiogram was performed before death. The heart was ex-
cised and the dimensions (cm²) of the infarcted area were assessed. Tissue samples for histological examination were taken from the infarcted region (clearly demarcated thin-walled area), fixed in 4% paraformaldehyde, sectioned, and stained with hematoxylin and eosin.

Statistics. Results are expressed as means ± SE. Statistical analysis was performed by repeated measures analysis of variance (ANOVA) using BMDP P2V. Control data were analyzed separately in a one-way ANOVA to test for changes over time. Each variable was analyzed in a two-way ANOVA that produced treatment (nonligated vs. ligated) and time interactions. Where significant differences were identified by ANOVA, Fisher’s protected least significant difference tests were used to identify time points significantly different from control. Statistical significance was assumed to be present when P < 0.05. Regression analyses were performed using the general linear model approach.

RESULTS

Significant changes over time were evident in many of the sham-ligation control hemodynamic and hormonal measurements, with the majority of these changes of short duration and related to anesthesia and/or surgery. Significant and transient increases in plasma levels of creatine kinase (P < 0.001), ANP (P < 0.001), BNP (P < 0.001), NT-BNP (P < 0.001), endothelin (P < 0.001), plasma renin aldosterone (PRA; P < 0.001), and aldosterone (P < 0.001) were observed in the 6- to 12-h postanesthesia/surgery period. Plasma epinephrine concentrations were reduced during anesthesia and rose abruptly in the 4-h postanesthetic period, before returning to baseline levels (see Figs. 2–4). These changes occurred in association with a fall in mean arterial pressure (P < 0.001) and rise in pulmonary arterial pressure (P < 0.001) that lasted a similar 6–12 h (Fig. 1). Heart rate (P < 0.001) also increased promptly postanesthesia/surgery and slowly returned to baseline levels over the following 1–2 wk, whereas hematocrit fell gradually over the first 7 days before rising (P < 0.001; Table 1). Plasma norepinephrine levels (P < 0.01) tended to rise over the 5-wk study period (see Fig. 4).
Ligation of the homonymous and second diagonal coronary arteries resulted in electrocardiograph changes characteristic of myocardial injury, with marked S-T elevation. Enzymatic markers of myocardial necrosis, creatine kinase (12 h, control 1,858 ± 322 units vs. ligated, 3,470 ± 624 units, P < 0.01) and, more specifically, troponin T (24 h, 0.17 ± 0.01 vs. 8.65 ± 0.72 µg/l, P < 0.001) were significantly increased compared with control data. Left ventricular angiography revealed marked rounding and expansion of the left ventricular cavity at 5 wk postmyocardial infarction (PMI), with significant increases in both left ventricular end-diastolic volume (LVEDV; 85 ± 13 vs. 142 ± 19 ml, P < 0.001) and end-systolic volume (LVESV; 41 ± 4 vs. 102 ± 17 ml, P < 0.001) compared with controls. Left ventricular ejection fraction (LVEF) was also significantly decreased compared with control data (51 ± 2 vs. 30 ± 5%, P < 0.001) and was inversely related to infarct size (r = -0.87, P < 0.001). Contractile regions were limited to the anterior and posterior segments closest to the base of the heart, with pooling and delayed clearing of contrast medium in the infarcted apex a characteristic of all postinfarct ventriculograms. Postmortem macroscopic examination of the hearts at 5 wk PMI revealed well-defined anteroapical infarcts measuring 33.6 ± 5.0 cm² with marked transmural thinning compared with control hearts (12.0 ± 0.8 vs. 3.5 ± 0.4 mm, P < 0.001). In every case the full thickness of the free left ventricular wall was affected. Microscopically, the reorganization of the infarcted tissue was advanced by 5 wk. Both endocardial and epicardial aspects of the infarct were entirely fibrotic with a medial layer of residual, enucleated cardiomyocytes. The fibrous tissue consisted of invading fibroblasts with some collagen deposition and formation of granulation tissue.

Serial hemodynamic data are shown in Fig. 1 and Table 1. Coronary artery ligation induced a marked and immediate fall in cardiac output compared with sham-ligation controls, reaching a nadir at 6 h PMI (6.3 ± 0.6 vs. 3.5 ± 0.3 l/min, P < 0.001). Cardiac output gradually improved over the following 3 days, but was still significantly reduced at 5 wk (6.3 ± 0.2 vs. 5.1 ± 0.2 l/min, P < 0.01; Fig. 1). The decline in cardiac function was associated with a progressive reduction in mean arterial pressure (5 wk PMI, 93 ± 2 vs. 79 ± 3 mmHg, P < 0.001; Fig. 1), whereas heart rate was elevated relative to controls on days 5 and 7 PMI (P < 0.05; Table 1). Total peripheral resistance showed a short-term transient rise, peaking at 12 h PMI (12.6 ± 1.1 vs. 25.3 ± 2.0 mmHg·l⁻¹·min⁻¹, P < 0.001; Table 1). Coronary ligation also induced a rise in cardiac preload. This was documented by prompt and sustained but moderate increases in right atrial (5 wk PMI, 1.6 ± 0.2 vs. 4.9 ± 0.9 mmHg, P < 0.01), mean pulmonary arterial (16.8 ± 0.2 vs. 18.9 ± 1.2 mmHg, P < 0.01), and left atrial pressures (3.3 ± 0.1 vs. 8.3 ± 1.3 mmHg, P < 0.01; Fig. 1 and Table 1). An increase in plasma volume was suggested by reductions in hematocrit from week 2 onward compared with control (P < 0.01; Table 1).

Serial hormone data are shown in Figs. 2–4 and Table 2. Coronary ligation significantly increased plasma BNP concentrations compared with control data at 6 h PMI (7.5 ± 0.7 vs. 11.5 ± 1.7 pmol/l, P < 0.01), NT-BNP at 12 h PMI (46 ± 16 vs. 69 ± 21 pmol/l, P < 0.01), and ANP at 2 days PMI (14.3 ± 1.9 vs. 20.4 ± 3.3 pmol/l, P < 0.05). Plasma natriuretic peptide levels remained significantly elevated over the following 5 wk (Fig. 2). Coronary sinus concentrations of ANP, BNP, and NT-BNP measured up to day 7 (in control and ligated animals) showed similar patterns to levels in left atrial blood (above), although in the coronary sinus, absolute levels of all peptides were markedly increased (Table 2). Plasma endothelin levels were only moderately elevated overall compared with control data after coronary artery ligation (P < 0.05; Fig. 3), and were not elevated at 5 wk PMI. Endothelin concentrations measured in coronary sinus samples were not significantly different from those found in left atrial samples (Fig. 3). Plasma epinephrine levels also tended to be elevated compared with control data (P < 0.05), although again, this was not true at 5 wk PMI (Fig. 4). Plasma renin and aldosterone were remarkably stable and showed no significant differences from control levels over the 5-wk
study period. Norepinephrine was not significantly raised compared with controls (Fig. 4).

Significant negative correlations were observed between interangiographic changes in LVEF (ΔLVEF) and plasma BNP concentrations measured on days 1–7 and at 5 wk PMI (with r values ranging from −0.849, P < 0.001). Positive correlations occurred between plasma BNP and ΔLVESV (r = 0.853, P < 0.001) and ΔLVEDV (r = 0.878, P < 0.001). Similarly, plasma NT-BNP levels measured on days 1–7 and at 5 wk PMI correlated negatively with ΔLVEF (r = −0.686, P < 0.01) and positively with ΔLVESV (r = 0.696, P < 0.01). Changes in ejection fraction and ventricular volumes did not show significant correlations with plasma ANP levels measured on days 1–5 PMI, but did correlate with concentrations sampled on day 7 (ΔLVEF: r = −0.684, P < 0.01; ΔLVESV: r = 0.713, P < 0.01) and 5 wk PMI (ΔLVEF: r = −0.644, P < 0.05; ΔLVESV: r = 0.782, P < 0.01). Changes in ejection fraction and ventricular volumes did not correlate significantly with any other hormones measured.

**DISCUSSION**

This study demonstrates that ligation of the left anterior descending artery and its second diagonal branch in sheep results in reproducible anteroapical

### Table 2. Coronary sinus natriuretic peptide responses to coronary artery ligation in 5 sheep

<table>
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<tr>
<th></th>
<th>Preligation</th>
<th>12 h Postligation</th>
<th>1 Day PMI</th>
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<th>3 Days PMI</th>
<th>7 Days PMI</th>
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<tr>
<td>Control</td>
<td>194 ± 23</td>
<td>172 ± 29</td>
<td>124 ± 15</td>
<td>86 ± 9</td>
<td>77 ± 4</td>
<td>79 ± 7</td>
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<td>Ligated</td>
<td>195 ± 32</td>
<td>218 ± 45</td>
<td>227 ± 54‡</td>
<td>181 ± 27‡</td>
<td>185 ± 40‡</td>
<td>208 ± 127‡</td>
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<tr>
<td>Control</td>
<td>41 ± 7</td>
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<td>38 ± 5</td>
<td>34 ± 7</td>
<td>34 ± 4</td>
<td>40 ± 8</td>
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<tr>
<td>Ligated</td>
<td>53 ± 5</td>
<td>111 ± 12‡</td>
<td>221 ± 24‡</td>
<td>109 ± 21‡</td>
<td>114 ± 23‡</td>
<td>130 ± 32‡</td>
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<td><strong>NT-BNP, pmol/l</strong></td>
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<tr>
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<td>37 ± 5</td>
<td>88 ± 26</td>
<td>43 ± 6</td>
<td>29 ± 5</td>
<td>26 ± 5</td>
<td>38 ± 10</td>
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<tr>
<td>Ligated</td>
<td>81 ± 17</td>
<td>220 ± 19‡</td>
<td>196 ± 32‡</td>
<td>192 ± 48‡</td>
<td>191 ± 59‡</td>
<td>147 ± 42‡</td>
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Coronary sinus (CS) levels of atrial natriuretic peptide (ANP), brain natriuretic peptide (BNP), and amino-terminal (NT) BNP after coronary artery ligation in 5 sheep compared with 3 sham controls. Values shown are means ± SE. Significant differences between control and ligated data are shown by ‡P < 0.001.
infarcts with ensuing left ventricular remodeling and dysfunction. Chronic hemodynamic changes included mild to moderate reductions in ejection fraction, cardiac output, and mean arterial pressure and increases in cardiac preload. These changes were associated with acute and sustained increases in plasma concentrations of ANP, BNP, and NT-BNP. Plasma levels of endothelin-1, renin activity, aldosterone, norepinephrine, and epinephrine showed little if any chronic elevation (5 wk) postinfarction compared with control data. Of all the hormones measured, only the natriuretic peptides (particularly BNP and NT-BNP) correlated significantly with indicators of left ventricular function (ejection fraction and left ventricular volumes). In contrast to the natriuretic peptides, endothelin concentrations were not raised in coronary sinus samples relative to peripheral samples.

This ovine model of left ventricular infarction and chronic left ventricular failure has a number of significant advantages over other experimental animal models currently used in the study of heart failure. First, and most importantly, the underlying pathology is comparable to that commonly found in the human situation, unlike other models based on pharmacologically induced myocardial damage where toxicity may be systemic (16, 34) or in pacing-induced heart failure where heart rate is fixed and the resulting ventricular remodeling is less typical (10, 21). Second, sheep have a consistent human-type coronary circulation (without multiple collaterals) (22), thus coronary artery ligation in these animals results in predictable and uniform anterolateral myocardial infarcts. In contrast, the canine coronary ligation model of heart failure is limited for study of ischemic heart disease because of abundant subepicardial coronary collateral vessels (8), making formation of apical aneurysms unpredictable and inconsistent. Third, unlike smaller animals such as rats (32), sheep are large enough to facilitate chronic invasive hemodynamic measurements and allow repeated sampling of a wide range of hormones, which is essential to adequately determine hemodynamic and endocrine interrelationships. In addition, sheep are widely available, relatively inexpensive, resistant to infection, and adjust well to handling.

In agreement with the findings of Markovitz et al. (22), and consistent with clinical and postmortem observations of left ventricular aneurysms in humans (3), coronary ligation resulted in left ventricular anterolateral infarcts, which at 5 wk showed marked dilatation and transmural thinning of the infarcted region. In accord with the extent of cardiac injury, we observed early characteristic increases in plasma levels of enzymatic markers of myocardial necrosis, creatine kinase, and more specifically, troponin T. Functional as well as structural alterations or remodeling of the left ventricle were present that included prominent rounding and expansion of the left ventricular cavity (as evidenced by the significant increases in left ventricular volumes) in conjunction with marked reductions in wall motion and declines in LVEF. Ventricular remodeling, a major consequence of infarction (39), is thought to be the essential factor underlying the progression from asymptomatic ventricular dysfunction to overt clinical heart failure. The depressed left ventricular performance demonstrated by these animals was associated with chronic and stable hemodynamic changes, including significant reductions in cardiac output and arterial pressure and increases in cardiac preload. These hemodynamic changes are in accord with the findings of Millner et al. (23) who previously performed hemody-
namic investigation of this model and are consistent with those observed in human heart failure, supporting the comparability of this ovine model with the clinical situation.

It is striking that marked remodeling occurred in the presence of only mild to moderate hemodynamic abnormality and without activation of the circulating renin-angiotensin-aldosterone system (RAAS) or increase in circulating catecholamines. In contrast, natriuretic peptide levels were activated in a sustained fashion. This model offers the opportunity to address the role of the natriuretic peptides in remodeling (through augmentation or antagonism), where it may be of benefit (by promoting vasodilation, natriuresis, and RAAS suppression) (9). Similarly, the effects of antagonizing the RAAS in this model will be of major interest. Although circulating levels are not elevated, the cardiac tissue RAAS may well participate in remodeling. Other experimental interventions (for example, cytokine antagonism or endothelin blockers) may also be tested in this system, which will allow definition of effects on hemodynamics, circulating neurohumoral status, and remodeling.

The sustained increases in plasma concentrations of both ANP and BNP observed after coronary ligation in the present study are in agreement with previous reports of prolonged activation of the cardiac natriuretic peptide system in patients who have experienced (at least) medium-sized infarcts (5, 37). We also noted earlier elevation of plasma BNP levels PMI above control (at 6 h) compared with ANP (at 2 days), which is consistent with the more rapid induction of the gene expression of this peptide in ventricular tissue after infarction (36). In contrast to other hormones measured during this study, we found strong correlations between plasma ANP and BNP levels and changes in left ventricular ejection fraction and volumes, which concurs with the findings of a number of clinical studies showing the natriuretic peptides to be superior neurohumoral markers of cardiac impairment after myocardial infarction (1, 11, 24, 28, 33). In common with a number of recent reports we noted closer statistical...
associations for plasma BNP levels compared with ANP with ejection fraction (24, 33), particularly in the first few days PMI (2, 28). Furthermore, the relationship between ejection fraction and BNP was evident with concentrations measured both early and at 5 wk PMI, suggesting that early postinfarction BNP can aid in the prediction of left ventricular function chronically postinfection. These data are consistent with both the site of synthesis and the regulation of the cardiac natriuretic peptides. Plasma concentrations of ANP (normally released from the atrium in response to stretch and also from the ventricle in situations of cardiac overload) (9), and particularly BNP (predominantly secreted constitutively from the ventricle in response to ventricular wall stress) (17), appear to reflect the extent of myocardial damage and dysfunction and provide a prognostic index of cardiac function. These findings further confirm the comparability of this ovine model with the clinical condition.

Because of the recent development in the laboratory of Espiner and colleagues (31) of an assay for ovine NT-BNP (31), we were able to detail for the first time the serial response of this peptide after myocardial infarction and progression to mild heart failure. The amino-terminal portion of proBNP (NT-BNP) circulates in human (15) and ovine (31) plasma, and levels are elevated in patients with chronic left ventricular dysfunction (14) and acutely following myocardial infarction (33). In this study we found the pattern of plasma NT-BNP to closely parallel that of (carboxy-terminal) BNP. The markedly higher plasma concentrations of NT-BNP compared with BNP probably reflect slower degradation (14, 31). The relationships between NT-BNP and LVEF and LVESV were comparable with those observed for BNP (both acutely and at 5 wk PMI). These findings concur with those reported in humans with cardiac impairment (14) and acutely following myocardial infarction (33) and are consistent with the view that NT-BNP is secreted together with BNP from the heart (41). Accordingly, we observed a significant step-up in natriuretic peptide concentrations in plasma taken from the coronary sinus compared with the left atrium. The current findings indicate NT-BNP to be at least as good a marker of cardiac dysfunction as BNP. Indeed, two recent studies in humans have reported that, for a given degree of cardiac injury, absolute and proportional increments in NT-BNP exceed those in BNP (14, 33), suggesting that it may be the more sensitive marker.

In contrast to the natriuretic peptides, no other hormones measured in this study were chronically and clearly elevated PMI. These data are consistent with studies in patients who have experienced smaller infarcts, which suggests that plasma concentrations of renin, angiotensin II, and aldosterone rise only slightly before returning to normal during the first days postinfarction (5). It is pertinent to speculate that the rise in circulating natriuretic peptides may act to suppress the RAAS and contribute to the pathophysiological compensation of early left ventricular dysfunction. This concept is supported by studies in experimental heart failure in which acute activation of ANP served to inhibit activation of the RAAS and maintain sodium balance despite the stimulation of arterial hypotension (19). In addition, Lohmeier et al. (20) demonstrated in dogs with pacing-induced heart failure that atrial appendectomy resulted in reduced plasma ANP levels and increased activation of the RAAS. This study also found increased levels of norepinephrine after atrial appendectomy compared with controls, which is in keeping with the inhibitory actions of the natriuretic peptides on sympathetic nervous activity (13), and may in part explain the failure of norepinephrine to rise significantly in this study. An additional contributing factor may be that, although myocardial infarction is painful and provokes intense anxiety, both of which are potent stimuli to sympathetic activation, the animals in this study were anesthetized during infarction and received analgesia immediately postoperatively. Furthermore, several clinical studies have reported marked sympathetic activation to be associated with large infarcts and a tendency to cardiogenic shock, arrhythmias, and death (4, 27), which were not features of the current study. From the significant alterations in hemodynamic and hormonal indexes demonstrated by the sham-ligated animals during anesthesia/surgery, this study exemplifies the importance of appropriate controls.

Plasma endothelin-1 concentrations PMI tended to be higher overall compared with control levels, however, this was not marked. Although there have been repeated demonstrations of raised circulating endothelin acutely following myocardial infarction (29) and in heart failure (6), there is increasing evidence that plasma endothelin is significantly raised only in moderate or severe heart failure (NYHA III and IV) not in mild (NYHA I) (40). This is consistent with the findings in the present study and, as may be the case with the RAAS, the elevated levels of the natriuretic peptides may be acting to suppress this system also (18). Although we observed a significant step-up in natriuretic peptide concentrations in plasma taken from the coronary sinus compared with the left atrium, we found no difference in endothelin levels between these two sites, suggesting the heart is not a major source of circulating endothelin postinfarction. This is in agreement with a study by Wei et al. (40) showing that although immunoreactive endothelin is present in normal atrial and ventricular tissue, there is no change in immunoreactivity even in severe heart failure. Clearly, the current model offers the opportunity to further probe these potential effects and inter-relationships.

In conclusion, this study documents the acute and chronic neurohormonal profile of a previously reported ovine model of acute myocardial infarction and left ventricular dysfunction. We demonstrate that coronary artery ligation induces acute and sustained elevations in circulating levels of the cardiac natriuretic peptides. We report for the first time serial plasma NT-BNP concentrations following myocardial infarction, and we show the pattern of secretion to closely follow that of BNP. Of the hormones measured, the natriuretic pep-
tides, particularly BNP and NT-BNP, best reflect the extent of myocardial damage and left ventricular dysfunction. This is a reproducible model that reflects the pathological, hemodynamic, and hormonal features of postmyocardial infarction remodeling and should allow future study of the progression of the disease as well as investigation of experimental therapeutic interventions on the natural history of ischemic left ventricular impairment.

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