Reproducibility of cardiac power output and other cardiopulmonary exercise indices in patients with chronic heart failure

Djordje G. JAKOVLJEVIC∗†, Petar M. SEFEROVIC‡, David NUNAN†, Gay DONOVAN†, Michael I. TRENELL∗, Richard GROCOTT-MASON§ and David A. BRODIE†

∗Institute for Ageing and Health, Faculty of Medicine, Newcastle University, Newcastle upon Tyne NE2 4HH, U.K., †Research Centre for Society and Health, Buckinghamshire New University, Uxbridge UB8 1NA, U.K., ‡Department of Cardiology, Clinical Centre of Serbia, Belgrade 11000, Serbia, and §Department of Cardiology, Hillingdon Hospital NHS Trust, Uxbridge UB8 3NN, U.K.

ABSTRACT

Cardiac power output is a direct measure of overall cardiac function that integrates both flow- and pressure-generating capacities of the heart. The present study assessed the reproducibility of cardiac power output and other more commonly reported cardiopulmonary exercise variables in patients with chronic heart failure. Metabolic, ventilatory and non-invasive (inert gas re-breathing) central haemodynamic measurements were undertaken at rest and near-maximal exercise of the modified Bruce protocol in 19 patients with stable chronic heart failure. The same procedure was repeated 7 days later to assess reproducibility. Cardiac power output was calculated as the product of cardiac output and mean arterial pressure. Resting central haemodynamic variables demonstrate low CV (coefficient of variation) (ranging from 3.4% for cardiac output and 5.6% for heart rate). The CV for resting metabolic and ventilatory measurements ranged from 8.2% for respiratory exchange ratio and 14.2% for absolute values of oxygen consumption. The CV of anaerobic threshold, peak oxygen consumption, carbon dioxide production and respiratory exchange ratio ranged from 3.8% (for anaerobic threshold) to 6.4% (for relative peak oxygen consumption), with minute ventilation having a CV of 11.1%. Near-maximal exercise cardiac power output and cardiac output had CVs of 4.1 and 2.2%, respectively. Cardiac power output demonstrates good reproducibility suggesting that there is no need for performing more than one cardiopulmonary exercise test. As a direct measure of cardiac function (dysfunction) and an excellent prognostic marker, it is strongly advised in the assessment of patients with chronic heart failure undergoing cardiopulmonary exercise testing.

INTRODUCTION

Cardiopulmonary exercise testing data have been widely used in cardiac patients to assess functional capacity and to determine prognosis. Repeated measurements obtained during cardiopulmonary exercise tests have generally shown good reproducibility in healthy adults [1] and in highly trained athletes [2]. A basic requirement for the clinical application of exercise-derived cardiopulmonary indices is high reproducibility.

Key words: cardiac function, cardiac power output, exercise testing, heart failure, reproducibility.

Abbreviations: CPO, cardiac power output; CV, coefficient of variance; DBP, diastolic blood pressure; MAP, mean arterial pressure; QT, cardiac output; RER, respiratory exchange ratio; SBP, systolic blood pressure; Vco2, carbon dioxide production; Ve, minute ventilation; VO2, oxygen consumption; VpeakO2, peak VO2.

† Correspondence: Dr Djordje Jakovljevic (email d.jakovljevic@ncl.ac.uk).
Particularly in patients with chronic heart failure, high reproducibility is of great importance because even small changes in cardiopulmonary data due to treatment or disease progression may have clinical significance [3].

Several studies have assessed reproducibility of cardiopulmonary exercise variables in patients with chronic heart failure [3–9]. Reproducibility of VO2peak [peak VO2 (oxygen consumption)] has been most frequently analysed. This is unsurprising, as in addition to reflecting physiological reserve of the cardiovascular system, VO2peak plays an important role in risk stratification, management and evaluation of the efficacy of treatment in patients with heart failure.

The importance of evaluating exercise haemodynamics in patients with heart failure has been previously described [10–12]. CPO (cardiac power output) is a central haemodynamic measure that demonstrates overall cardiac function and is the product of simultaneously measured QT (cardiac output) and MAP (mean arterial pressure) [13,14]. By incorporating both pressure and flow domains of the cardiovascular system, CPO is a unique, direct and integrative measure of cardiac pumping capability [13,15,16]. Not surprisingly, resting and peak exercise CPO have been shown to be powerful predictors of prognosis and mortality in patients with chronic heart failure [17–21]. Recently, our group has shown that CPO differentiates well during cardiac restoration using left ventricular assist devices [22] and emphasizes the benefits of this therapy in terms of organ function at rest and peak exercise [23]. It has also been shown that commonly reported cardiopulmonary exercise-derived indices demonstrate limited capacity in reflecting cardiac pumping capability in patients treated with ventricular assist devices [24].

In contrast with other, well-established cardiopulmonary exercise indices (e.g. VO2), reproducibility of CPO has been reported by one study only, using a specific two-stage exercise protocol [25]. However, reproducibility of CPO using a standard incremental exercise test has not yet been demonstrated. Therefore, the purpose of the present study was to assess the reproducibility of CPO in conjunction with other important cardiopulmonary exercise indices. We hypothesize that CPO will demonstrate a high degree of reproducibility in patients with chronic heart failure.

### MATERIALS AND METHODS

#### Patients

The study group consisted of 19 stable chronic heart failure patients with New York Heart Association classes I and II. The patients’ demographic and clinical characteristics are presented in Table 1. All patients were symptomatically stable and on the same dose of optimal drug therapy for the preceding 2 months. Exclusion criteria were exercise-induced myocardial ischaemia or uncontrolled arrhythmias, valvular heart disease, congestive heart failure, uncontrolled hypertension (resting SBP (systolic blood pressure) above 160 mmHg, and/or resting DBP (diastolic blood pressure) above 105 mmHg), peripheral vascular disease, chronic obstructive pulmonary disease and orthopaedic or other condition precluding participation in exercise testing. The present study was approved by the Hounslow and Hillingdon NHS Research Ethics Committee. An informed written consent form was obtained from each patient prior the investigation.

### Study protocol and measurements

In order to assess reproducibility of CPO and selected cardiopulmonary exercise variables, patients visited the Cardiac Rehabilitation Department of Hillingdon Hospital twice within 1 week of each other. All equipments were calibrated prior to each exercise test according to the manufacturer’s recommendations. Measurements were performed at the same time of day under identical conditions and medication. Patients who were not familiar with the use of treadmill were asked to visit the laboratory a few days prior to their first test to complete a brief familiarization protocol. This was not a maximal exercise test but familiarization with walking on a treadmill at different speeds.

All patients attended the clinic at least 2 h after having any food or drink and did not consume alcohol or caffeine.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>62 ± 11</td>
</tr>
<tr>
<td>Men/women (n)</td>
<td>15/4</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>79.6 ± 17</td>
</tr>
<tr>
<td>Height (cm)</td>
<td>172 ± 10</td>
</tr>
<tr>
<td>BMI (kg/m2)</td>
<td>26.6 ± 4.0</td>
</tr>
<tr>
<td>BSA (m2)</td>
<td>1.92 ± 0.25</td>
</tr>
<tr>
<td>Cause of heart failure (ischaemic/non-ischaemic) (n)</td>
<td>13/6</td>
</tr>
<tr>
<td>LVEF (%)</td>
<td>29 ± 12</td>
</tr>
<tr>
<td>Medication (n)</td>
<td></td>
</tr>
<tr>
<td>ACE inhibitors</td>
<td>12</td>
</tr>
<tr>
<td>β-Blockers</td>
<td>16</td>
</tr>
<tr>
<td>Diuretics</td>
<td>8</td>
</tr>
<tr>
<td>ARBs antagonists</td>
<td>6</td>
</tr>
<tr>
<td>Digoxin</td>
<td>5</td>
</tr>
<tr>
<td>Anti-arrhythmics</td>
<td>7</td>
</tr>
<tr>
<td>Aspirin</td>
<td>9</td>
</tr>
<tr>
<td>Warfarin</td>
<td>5</td>
</tr>
<tr>
<td>Spironolactone</td>
<td>6</td>
</tr>
</tbody>
</table>

Table 1 Patients’ demographic and clinical characteristics (n = 19)

ACE, angiotensin-converting enzyme; BMI, body mass index; BSA, body surface area; LVEF, left ventricular ejection fraction; ARBs, angiotensin II receptors blockers.
containing beverages before the tests. The tests were performed at an ambient room temperature maintained at approx. 20°C.

**System configurations**
Gas-exchange measurements were recorded on a breath-by-breath basis using the Cardio O₂ system (Medical Graphics), whereas \( Q_T \) was estimated using inert gas re-breathing methodology (Innocor; Innovision), as described below. The Innocor respiratory valve unit, with bacterial filter, was inserted into the pneumotach of the Medical Graphics system. Both systems were attached to the subject’s face mask, where the pneumotach of the Medical Graphics system was closer to the subject’s mouth. Such configuration increased dead space and the distance between the subject’s mouth and the Innocor gas sensors. The volume calibration was performed after which the two systems were placed in series. The Innocor respiratory valve unit was supported with the Innocor metal arm attached to the monitor trolley. Prior to measurements, all patients were familiarized with the face mask and a learning session of the Innocor \( Q_T \) procedure was performed.

**Resting measurements**
With the patient seated and following 5 min of acclimatization, arterial blood pressure was assessed from the brachial artery by cuff sphygmomanometry. Expired air was collected for online measurement of ventilation and expired gas concentrations. \( Q_T \) was measured using the inert gas re-breathing methodology while the patient was still in a seated position, as previously described [26–28]. Briefly, an oxygen-enriched mixture of an inert soluble gas (0.5 % nitrous oxide) and an inert insoluble gas (0.1 % sulfur hexafluoride) from a 4-litre pre-filled anaesthesia bag was used. Photo-acoustic analysers measure gas concentrations over a 5-breath interval. Nitrous oxide concentration decreases during the re-breathing manoeuvre at a rate proportional to pulmonary gas-exchange. \( \text{PetO}_2 \) fell by approx. 0.1 mmHg per minute of re-breathing. Three to four respiratory cycles were needed to obtain a value for nitrous oxide washout.

**Exercise measurements**
Patients then performed a modified Bruce protocol on a motor driven treadmill (Cardio Control). During the test, ventilation and expired gases were collected and analysed as described at rest. Arterial blood pressure and Borg scale recordings for dyspnoea and fatigue were performed during the last 30 s of each exercise stage. At near-maximal exercise [defined as at least one of the following: RER (respiratory exchange ratio) > 1.05, the absence of a rise in \( \text{V}O_2 \) with further increase in exercise intensity, Borg scale >17], \( Q_T \) was measured again using the re-breathing technique described at rest and blood pressure was recorded. Patients were instructed to give an approx. 30-s warning before they felt they would end the exercise so that a final \( Q_T \) re-breathing measurement was obtained at near-maximal exercise. Patients were verbally encouraged to continue exercise test for as long as they were able, despite termination of \( Q_T \) measurement. The time between near-maximal \( Q_T \) measurement and termination of exercise test was recorded. \( \text{VO}_2 \text{peak} \) was defined as the average \( \text{VO}_2 \) during the last minute of exercise. Anaerobic threshold was identified by the V-slope method [29].

**Calculations and statistical analysis**
The CPO was calculated from the product of \( Q_T \) and MAP using the following equation [20]:

\[
\text{CPO (Watts)} = (Q_T \times \text{MAP}) \times K
\]

where \( K \) is the conversion factor (2.22 × 10⁻³), \( Q_T \) is measured in l/min and MAP is in mmHg. Near-maximal exercise arterial-venous oxygen difference, expressed in ml of \( \text{O}_2 \)/100 ml of blood, was calculated as the ratio between \( \text{VO}_2 \text{peak} \) and peak \( Q_T \).

All statistical analysis was carried out using SPSS version 13.0. Prior to statistical analysis, data were checked for univariate and multivariate outliers using standard Z-distribution cut-offs and Mahalanobis distance tests respectively. Normality of distribution was assessed using a Kolmogorov–Smirnov test. To test differences in measured variables between test I and test II, a Student’s \( t \) test for paired samples was used. Statistical significance was indicated if \( P < 0.05 \). To assess the reproducibility of CPO and selected cardiopulmonary variables, the CV (coefficient of variation) and Pearson’s correlation coefficient (\( \rho \)) were calculated. For each variable, the CV was defined as the (within-person S.D./within-person mean) × 100 %. A CV of ≤ 6 % was considered as an indicator of good, 6–10 % acceptable, and >10 % poor reproducibility. Additionally, Bland–Altman analysis was used to demonstrate between-test agreement of cardiopulmonary exercise test variables measured at peak exercise [30]. All data are presented as means ± S.D. unless otherwise indicated.

**RESULTS**
All subjects completed each exercise test without any contraindications. Thirteen patients stopped the first exercise test due to fatigue and six patients stopped due to dyspnoea. Four patients reported a different limiting symptom at the end of the second test. Time between \( Q_T \) measurement and termination of exercise test was 16 ± 8 for test 1 and 22 ± 11 min for test 2 (\( P > 0.05 \)).

There were no significant differences in resting metabolic, ventilatory and central haemodynamic variables between the two tests (Table 2). Central haemodynamic
Reproducibility of resting measurements in patients with chronic heart failure

DISCUSSION

In the present study, we evaluated the reproducibility of CPO, a direct measure of cardiac function and pumping capability [13,14], and other important cardiopulmonary exercise parameters in patients with chronic heart failure. The major findings suggest that (i) CPO measured at rest and near-maximal exercise demonstrates good reproducibility and that (ii) exercise duration, $V\text{CO}_2$ and $V\text{E}$ also demonstrate low CV.

A common method of assuring a reproducible response to exercise is to have the patient perform two exercise tests on separate days, at the same time of the day, and a test is considered reproducible if peak oxygen uptake is within $\pm 10\%$ on both days [31]. Reproducible physiological responses to exercise are obtainable over extended periods of time in patients with stable chronic heart failure [32]. In the light of this finding and according to recent reproducibility studies [4,5], the present study was designed to assess reproducibility of CPO in patients with stable mild chronic heart failure performing two exercise tests within 1 week of each other.

CPO is emerging as an important physiological variable in the assessment of cardiac function and prognosis in heart failure patients. Therefore the findings of the present study are discussed from the following perspectives: (i) exercise physiology, (ii) prognostic utility and (iii) feasibility of measurement.

Central haemodynamics and CPO demonstrate low CV and good reproducibility. An increase in exercise duration of 61 s, and consequent increase in metabolic and ventilatory responses, suggest that patients were able...
to demonstrate greater effort during the second test. These findings are likely to be explained by 'learning effect', previously demonstrated in heart failure patients [7], and also perhaps due to day-to-day variability in physiological variables described earlier [33]. Increased $V_{O_2}$ peak on the second test was due to a 4% increase in arterial-venous oxygen difference as $Q_T$ was not different between the two tests. This is an important finding because it suggests that cardiac function and pumping capability are less affected by exercise test 'learning effect' and physiological day-to-day variation observed in gas exchange measurements. The assessment of CPO does not require performing more than one cardiopulmonary exercise test.

The present results demonstrate, for the first time, good reproducibility of resting CPO in chronic heart failure patients, having a CV of 5% and correlation coefficient of 0.96. This is an important finding as in contrast with resting metabolic and ventilatory variables, resting CPO may have a practical implication in the detection of therapy-derived changes in cardiac function and prognosis in patients with heart failure [17,18]. $Q_T$ measured at rest by the inert gas re-breathing methodology (a component of CPO) is also highly reproducible, in agreement with previous reports [26,34], and demonstrates a CV below 10% in heart failure patients. In contrast with central haemodynamics, resting gas exchange measurements demonstrated greater CV. There are many uncontrollable factors that could affect reproducibility of resting metabolic measurements and most are beyond the control of the study. Although all patients were instructed to eat nothing at least 2 h prior to the exercise tests and not to consume alcohol or caffeine-containing beverages before the tests, there is no guarantee that this was indeed the case. Other causes may include variations in the amount of sleep, degree of activity during the previous day, dietary salt load and fluid intake, and emotional state.

The results further reveal good reproducibility of near-maximal exercise CPO demonstrated by a low CV (4.1%), high correlation coefficient ($r = 0.96$), small mean difference ($-0.02$ W) and narrow limits of agreement ($-0.14$ to $0.10$ W) between repeated measures. This should be unsurprising as both components of CPO (i.e. $Q_T$ and MAP) demonstrate good reproducibility at peak exercise. To date, only one study has reported the reproducibility of exercise cardiac power in patients with heart failure [25]. In that study, patients performed a treadmill exercise test in two stages with a 40 min resting period between the two stages. The first stage was an incremental test to determine the maximum $Vco_2$, and a second maximum workload stage was used to make $Q_T$ measurements. Unlike that study, we deliberately selected a protocol which is commonly used in clinical assessment. Using different methodology, Cooke et al. [25] reported a CV for CPO of 9.1%. Although the authors [25] questioned the reproducibility of arterial blood pressure measurement by cuff manometry during exercise, they did not report repeated blood pressure measurements, or reproducibility analysis of this variable. However, the good reproducibility and low CV for blood pressure found in the present study has been reported elsewhere [3,7].

Unlike CPO, reproducibility of well-established cardiopulmonary exercise test variables and particularly $V_{O_2}$ are well documented in single- and multi-site

### Table 3  Reproducibility of cardiopulmonary exercise testing measurements in patients with chronic heart failure

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Test I</th>
<th>Test II</th>
<th>P value</th>
<th>$r$</th>
<th>CV (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Metabolics</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>$V_{O_2}$ (ml/min)</td>
<td>1792 ± 27</td>
<td>1804 ± 27</td>
<td>0.04</td>
<td>0.99</td>
<td>5.7</td>
</tr>
<tr>
<td>$V_{O_2}$ (kg of body weight per min)</td>
<td>21.7 ± 7.0</td>
<td>23.0 ± 5.8</td>
<td>0.03</td>
<td>0.97</td>
<td>6.4</td>
</tr>
<tr>
<td>$V_{O_2}$ at AT (kg of body weight per min)</td>
<td>13.7 ± 3.5</td>
<td>14.2 ± 3.4</td>
<td>0.14</td>
<td>0.97</td>
<td>3.8</td>
</tr>
<tr>
<td>$Vco_2$ (ml/min)</td>
<td>1953 ± 251</td>
<td>2147 ± 337</td>
<td>0.11</td>
<td>0.98</td>
<td>5.5</td>
</tr>
<tr>
<td>$V_{E}$ (l/min)</td>
<td>53.4 ± 16.9</td>
<td>60.2 ± 21.7</td>
<td>0.02</td>
<td>0.90</td>
<td>11.1</td>
</tr>
<tr>
<td>RER</td>
<td>1.09 ± 0.04</td>
<td>1.14 ± 0.08</td>
<td>0.13</td>
<td>0.91</td>
<td>4.2</td>
</tr>
<tr>
<td>Exercise time (s)</td>
<td>604 ± 191</td>
<td>745 ± 190</td>
<td>0.00</td>
<td>0.95</td>
<td>7.1</td>
</tr>
<tr>
<td>Borg score</td>
<td>16.8 ± 2.8</td>
<td>17.6 ± 2.1</td>
<td>0.18</td>
<td>0.91</td>
<td>3.2</td>
</tr>
<tr>
<td><strong>Haemodynamics</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>$Q_{T}$ (l/min)</td>
<td>11.2 ± 5.7</td>
<td>11.3 ± 5.6</td>
<td>0.84</td>
<td>0.99</td>
<td>2.2</td>
</tr>
<tr>
<td>HR (beats/min)</td>
<td>116 ± 31</td>
<td>118 ± 26</td>
<td>0.42</td>
<td>0.96</td>
<td>4.4</td>
</tr>
<tr>
<td>SBP (mmHg)</td>
<td>148 ± 27</td>
<td>146 ± 25</td>
<td>0.67</td>
<td>0.93</td>
<td>4.5</td>
</tr>
<tr>
<td>DBP (mmHg)</td>
<td>74 ± 13</td>
<td>78 ± 11</td>
<td>0.23</td>
<td>0.90</td>
<td>7.5</td>
</tr>
<tr>
<td>MAP (mmHg)</td>
<td>104 ± 17</td>
<td>106 ± 15</td>
<td>0.51</td>
<td>0.98</td>
<td>6.8</td>
</tr>
<tr>
<td>CPO (W)</td>
<td>2.68 ± 1.53</td>
<td>2.69 ± 1.52</td>
<td>0.63</td>
<td>0.96</td>
<td>4.1</td>
</tr>
</tbody>
</table>
found that although there was a significant improvement in exercise time, there was no significant change in $\text{VO}_2\text{peak}$. Similarly, as in the present study, others also reported high reproducibility for duplicate measurements of cardiopulmonary exercise testing variables in patients with heart failure. These studies report a CV for $\text{VO}_2\text{peak}$ of 3% and 9% [3, 6, 8]. In the largest trial (398 heart failure patients), Bensimhon et al. [5] reported $\text{VO}_2\text{peak}$ and anaerobic threshold to be unchanged from test 1 to test 2, with the CVs of 6.6 and 7.8% [5]. In contrast, mean exercise time increased significantly from test 1 to test 2 by 4%. Most recently, in a multi-site clinical trial setting Keteyian et al. [4] assessed reproducibility of $\text{VO}_2\text{peak}$ and anaerobic threshold and reported CVs of 5.9 and 6.8%. The present study confirms good reproducibility of $\text{VO}_2\text{peak}$ and anaerobic threshold. The difference of 5.7% (1.3 ml/kg of body weight per min) is unlikely to have practical and clinical implications and is likely to be an indication that patients were able to tolerate greater effort during the second test as a consequence of ‘learning effect’.

It has been shown that both resting and near-maximal exercise CPO are excellent predictors of prognosis in patients with heart failure [17–21]. Also CPO obtained at near-maximal exercise is a direct measure of overall cardiac function and pumping capability of the heart [13, 14]. The findings of the present study suggest that both resting and near-maximal exercise CPOs demonstrate good reproducibility. Therefore, it is reasonable to suggest that there is no need for performing more than one cardiopulmonary exercise test to assess CPO in patients with stable chronic heart failure.

Finally, it is important to mention feasibility of $Q_T$ and CPO measurement during exercise. Monitoring $Q_T$ at rest and particularly during exercise is not a straightforward procedure [12]. Gold standard methods such as direct Fick and thermodilution are invasive, expensive, require specialist expertise, and are associated with inherent risks. Several non-invasive methods for $Q_T$ assessment have been developed, such as ultrasound techniques, impedance cardiography, bioreactance and pulse contour analysis. However, validity and reliability of these techniques during exercise have often been questioned since they are sensitive to motion artefacts. On the other hand, re-breathing ($\text{CO}_2$ and inert gas) methods are, however, frequently used in both healthy and clinical populations, including heart failure patients. Re-breathing methods, however, require patients to learn the re-breathing technique, are effort dependent and provide discontinuous, single-point measurement. Also from our experience approx. 10–15% of patients are unable to perform the re-breathing technique, and this seems to be associated with severity of disease. Nonetheless, while waiting for an ‘ideal’ method to be developed, re-breathing techniques are appropriate for estimating $Q_T$ and CPO.

The major limitation of the present study is inclusion of patients with mild heart failure only, with well-preserved exercise capacity. It is possible that those with more advanced stages of heart failure with significantly lower levels of maximal exercise capacity may demonstrate an altered response of cardiac function to increasing exercise levels.

In conclusion, CPO demonstrates good reproducibility suggesting that there is no need for performing more than one cardiopulmonary exercise test. As a direct measure of cardiac function (dysfunction) and an excellent prognostic marker, it is strongly advised in the assessment of patients with chronic heart failure undergoing cardiopulmonary exercise testing. Future research is warranted to evaluate reproducibility of CPO in multi-site protocols.

**AUTHOR CONTRIBUTION**

Djordje Jakovljevic designed and conducted the study, and analysed data under the supervision of David Brodie and Richard Grocott-Mason. Petar Seferovic and Michael Trenell provided intellectual input. The study was conceived by David Nunan and Gay Donovan. All authors co-wrote the paper.

**ACKNOWLEDGEMENT**

We thank all of the patients for taking part in this study.

**FUNDING**

This work was funded by the Research Centre for Health at Buckinghamshire New University.

**REFERENCES**


Received 12 July 2011; accepted 1 September 2011 Published as Immediate Publication 1 September 2011, doi:10.1042/CS20110355

© The Authors Journal compilation © 2012 Biochemical Society