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## Pharmacokinetics of intravenous *N*-acetylcysteine in men at rest and during exercise

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**Abstract Objective:** We aimed to determine the pharmacokinetics (PK) of *N*-acetylcysteine (NAC) at rest and during exercise when given by continuous intravenous infusion intended to maintain relatively constant plasma concentrations.

**Methods:** Plasma concentrations of NAC were measured in 24 healthy male subjects during and after a two-stage intravenous infusion designed to provide constant NAC concentrations during cycling exercise, including intense exercise to fatigue.

**Results:** A three-compartment, open PK model was the best fit using population PK analysis with NONMEM. Whole-body clearance (CL) was  $0.58 \text{ l kg}^{-1} \text{ h}^{-1}$  (95% CI 0.44–0.72) for reduced NAC (NACR) and  $0.16$  ( $0.13$ – $0.20$ )  $\text{l kg}^{-1} \text{ h}^{-1}$  for total NAC (NACT). The central volume of distribution (V1) was  $0.064$  ( $0.008$ – $0.12$ )  $\text{l kg}^{-1}$  for NACR and  $0.037$  ( $0.02$ – $0.06$ )  $\text{l kg}^{-1}$  for NACT. Exercise was a significant covariate in the model, resulting in a 25 and 23% reduction in CL of NACR and NACT, respectively. V1 in our subjects was smaller than expected, resulting in higher-than-anticipated initial concentrations of NAC. Despite these findings, the incidence of adverse effects attributable to NAC was minimal without using prophylactic or concomitant drug therapy.

**Conclusions:** NAC can be given to healthy exercising men by intravenous infusion and to the plasma concentrations seen in this study with minimal adverse effects due to the drug. The PK parameters of NAC at rest in volunteers are consistent with previously reported values and are significantly altered by vigorous cycling exercise.

### Introduction

Antioxidant compounds have been used in many investigations of skeletal muscle fatigue. The use of *N*-acetylcysteine (NAC) to attenuate muscle fatigue in humans was first reported by Reid et al. [1], who also observed a number of significant adverse effects attributable to the drug. We have recently reported the effects of NAC on exercise and blood redox status [2–4] during short-term, intense, and prolonged cycling exercise in healthy men. In those studies, we aimed to infuse NAC to achieve a relatively constant plasma concentration, assuming that if the effects of NAC were concentration-dependent, the effects should also be relatively constant during the exercise phases. The target plasma concentration of NAC was chosen based on available data from published studies. In particular, we were assisted by the report of adverse effects [1] in a study that showed a steady increase in incidence of reactions to NAC infusion during infusion at a constant rate over 1 h.

A limited number of pharmacokinetic studies have been published in which NAC was given by intravenous infusion. Borgstrom et al. [5] infused 600 mg of NAC over 5 min, approximating a bolus dose, and observed rapidly changing plasma concentrations. Olsson et al. [6] gave 200 mg intravenously, also as a bolus, and measured both total and reduced NAC over 12 h in healthy subjects. This was the first report distinguishing the differing pharmacokinetics of NAC in both the reduced form and as total NAC. Prescott et al. [7] determined the pharmacokinetics of intravenous NAC in patients with

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paracetamol overdose. Although not all authors reported adverse effects during the use of NAC, those that did generally related their occurrence to higher concentrations or to times when the higher concentration would be expected [1, 8, 9]. The most common adverse effects, sometimes termed “anaphylactoid”, include flushing, urticaria, bronchospasm, pruritis, angioedema, and nausea and vomiting. On the assumption that histamine release may underlie some of these effects, the use of antihistamines has been recommended although we planned to avoid concomitant drug administration, choosing, instead, to treat adverse effects if and as they occurred. We had three main aims in this study: First, to design an infusion protocol that would achieve relatively constant plasma NAC concentrations during the exercise phase while avoiding adverse effects; second, to determine the pharmacokinetics of both reduced and total NAC in our subjects at rest and during exercise; and third, to either confirm our original protocol or revise it based on the pharmacokinetic analysis.

## Methods

The overall study design has been published previously [2, 3, 4]. Twenty-four men (age: mean 23.5, SD 4.5 years; body mass: mean 77.7, SD 9.9 kg; height: mean 179.1, SD 4.7 cm) volunteered for the study after being informed of all known risks and giving written informed consent. Subjects refrained from vigorous activity and avoided ingesting alcohol, caffeine, or other drugs in the 24 h before the studies. They were required to have no history of asthma, bronchospasm, or atopy; no previous exposure to NAC; and not to be taking medication at the time of the study. Women were specifically excluded. Eight subjects participated in a study of the effects of intermittent high-intensity exercise comprising three bouts of 45 s followed by a final bout to fatigue, each bout at a power output corresponding to 130% of  $\dot{V}O_{2peak}$ , as detailed previously [2]. Sixteen other subjects took part in studies of lower intensity but more prolonged exercise comprising 45 min at 70%  $\dot{V}O_{2peak}$  then to fatigue at 90%  $\dot{V}O_{2peak}$  [3, 4]. The protocol complies with the World Medical Association Declarations of Helsinki and revisions (2002) regarding ethical principles for medical research involving human subjects. Approval for the studies was obtained from the Victoria University of Technology Human Research Ethics Committee.

### Design of the infusion protocol

The aim was to achieve a target plasma concentration of NAC sufficient to produce a pharmacological effect while avoiding concentration-dependent adverse effects. Based on published data, it was assumed that there is a plasma concentration of NAC below which adverse effects are absent or minimal and above which frequency is either

dose- or concentration-dependent. The time course of cumulative adverse effects during a 1-h constant-rate infusion of NAC [1] is consistent with the time course of predicted plasma concentrations. Using pharmacokinetic data derived from prolonged NAC infusions in overdose patients [7], we simulated plasma NAC concentrations for the previously reported studies [1, 5, 6, 8, 9]. On the basis of this model, we estimated that Reid's [1] subjects could have been expected to have plasma NAC concentrations below 100 mg l<sup>-1</sup> for the first 15 min of the infusion used. Thereafter, we estimated that the NAC concentration would eventually exceed 100 mg l<sup>-1</sup>, peaking at approximately 350 mg l<sup>-1</sup> by 60 min.

The avoidance of adverse effects has to be counter-balanced by the need to achieve drug concentrations that would be expected to be effective in terms of the experimental outcomes. Unfortunately, there are no data on the minimally effective concentrations of NAC in experimental studies of muscle fatigue in intact humans. We therefore chose a target of 100 mg l<sup>-1</sup> as being sufficient to avoid adverse effects and having the expectation of pharmacological effectiveness.

The general theory and method of rapidly achieving and maintaining a desired plasma concentration of a drug by the use of consecutive constant-rate infusions has been described [10, 11]. An initial infusion is given to achieve the target concentration quickly with or without some overshoot, depending on the rate and duration of the infusion. One or more subsequent infusions are used to maintain a relatively constant plasma concentration. Estimates of pharmacokinetic parameters are required for this method, and we used the same parameter set as for our simulations. With linear pharmacokinetic models, plasma drug concentrations following any input are a function of the input and disposition functions [12]. That is, a pharmacokinetic model derived from plasma concentration–time data will be independent of the input apart from scaling.

Using simulations based on our PK model, we designed an infusion protocol of two consecutive constant-rate infusions, the first of 125 mg kg<sup>-1</sup> h<sup>-1</sup> for 15 min followed by 25 mg kg<sup>-1</sup> h<sup>-1</sup> with the subjects at rest for 35 min, thereafter continuing the infusion during exercise until this phase was terminated by subject fatigue [2–4]. *N*-acetylcysteine (Parvolex, Faulding Pharmaceuticals, Melbourne, Australia) was diluted with 0.9% sodium chloride injection to a concentration of either 80 or 120 mg ml<sup>-1</sup>. The solution was infused into a forearm vein via a 22-gauge canula (Terumo, Sydney, Australia) using a syringe pump (Graseby 3400, Graseby Medical, Watford, UK) programmed to deliver the two infusions sequentially. The infusion site was chosen to allow the subject to have relatively free use of both arms and to enable constant inspection of the infusion site for local drug-related effects. Each subject acted as his own control by receiving on a separate occasion a 0.9% sodium chloride injection infusion at the same rate in the placebo phase of the study. A different vein was used on each occasion.

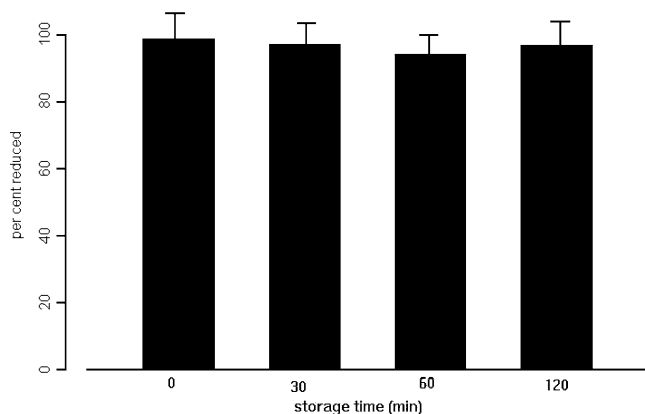
Arterialized venous blood [2] was withdrawn from the contralateral arm for determination of the concentrations of total and reduced thiols, plasma hemoglobin concentration, hematocrit and plasma concentrations of  $K^+$ ,  $Na^+$ ,  $Cl^-$ , and  $Ca^{2+}$  ions. In each subject, samples were taken before commencing the NAC infusion and then at 1, 2, 5, 10, 15, 25 and 35 min. In the intense exercise group, samples were taken at the beginning and end of each 45-s bout and at fatigue. Postfatigue samples were taken at 1, 2, 5, 10, 30, 60, 120 and 240 min. In the prolonged exercise group, samples were taken each 15 min during exercise and at fatigue. Postfatigue sampling was as for the previous study. Blood processing and analysis for total and reduced thiols have been detailed previously [2].

### Stability and form of the infused NAC solution

There are no data available on the relative amounts of NAC in the reduced or oxidized forms as infused or on the stability of these over time. This was determined by diluting the Parvolex to  $120 \text{ mg ml}^{-1}$  and storing the solution at room temperature for up to 120 min. At times 0, 30, 60 and 120 min, an aliquot was further diluted in phosphate-buffered saline, and ten samples of each were immediately assayed for both reduced NAC (NACR) and total NAC (NACT) using the method employed for the analysis of plasma. Difference between batches was tested using analysis of variance.

### Pharmacokinetic analysis

Concentration-time data for both NACR and NACT were modelled using the nonlinear mixed-effects program NONMEM [13]. The pharmacokinetics of NACR and NACT were modelled separately, and since the drug input was virtually entirely in the reduced form (Fig. 1), the same input data were used for each analysis. We



**Fig. 1** Stability of reduced *N*-acetylcysteine (NAC) after dilution of the Parvolex to  $120 \text{ mg ml}^{-1}$  with saline and storage in a syringe at room temperature for the time shown. Each bar shows the percent of the NAC in the reduced form and is the mean (SD) of ten samples

estimated population PK parameters with NONMEM. Although a pooled data approach could be used, particularly with subject groups with similar physical characteristics, the population method allows estimation of both between- and within-subject variability. This method assumes that there is a typical population value for each parameter and that each individual subject has a parameter value that differs from the population value by the between-subject variability, which can be estimated. Structural PK models were fitted to the concentration-time data using individual values, and all data were weighted equally. Both two- and three-compartment models were fitted to the data. The effect of addition or removal of a parameter was tested by the change in the reduction in the NONMEM objective function ( $-2 \times \log$  likelihood). Covariates such as exercise,  $\dot{V}O_2$ ,  $\dot{V}O_{2\text{peak}}$  and age were added to the model or removed according to the reduction in the objective function. A reduction of the objective function by 4 or more with inclusion was accepted as an improved fit.

Between-subject variability was modelled as an exponential error model.

$$\theta_i = TV\theta_i \cdot \exp(\eta_i), \quad (1)$$

in which  $\theta_i$  is the value of the parameter  $\theta$  in the  $i$ th subject,  $TV\theta$  is the typical or mean population value of this parameter, and  $\eta_i$  is a random variable with mean zero and variance  $\omega_\eta^2$ .

The remaining variability, which includes within-subject variability and measurement errors, was modelled using a proportional error model

$$C_{\text{meas},ij} = C_{\text{pred},ij} \cdot (1 + \varepsilon_{ij}), \quad (2)$$

where  $C_{\text{meas},ij}$  is the  $j$ th measured concentration in the  $i$ th individual,  $C_{\text{pred},ij}$  is the model prediction for that value, and  $\varepsilon_{ij}$  is a random variable with mean zero and variance  $\sigma_\varepsilon^2$ . NONMEM can provide estimates for the typical population parameter values, the between-individual variances, and the within-individual variances. The NONMEM subroutines ADVAN3 and ADVAN 11 with TRANS4 were used for building two- and three-compartment models, respectively. This reparameterization allowed the model parameters to be included as volumes of distribution, whole-body clearance, and intercompartmental clearances. The NONMEM first-order conditional estimates method (FOCE) was used.

Further information on the accuracy of a model can be obtained by calculating the median weighted residuals (MWR) and the median absolute weighted residuals (MAWR) as:

$$\text{MWR} = \text{median} [(C_{\text{meas},ij} - C_{\text{pred},ij})/C_{\text{pred},ij}], \quad (3)$$

$$\text{MAWR} = \text{median} \{ \text{ABS}[(C_{\text{meas},ij} - C_{\text{pred},ij})/C_{\text{pred},ij}] \}, \quad (4)$$

with MWR as a measure of bias and MAWR as a measure of precision.

Results are presented as the typical population values for the PK parameters with their between-subject variability. Body weight was not used as a covariate since all drug was infused on a per kilogram body-weight basis. Weight was included in the model as a scaling factor. As a covariate, the effect of exercise on parameters was examined both as a two-point proportional effect between the two levels of high intensity and the lesser prolonged intensity [2–4] and also as a single additive effect.

## Results

### Stability and form of the infused NAC solution

As infused, the NAC in solution was almost entirely in the reduced form, with the mean value in the 40 samples being 96.5 (6.8)%. The change of NAC in the reduced form measured over time are shown in Fig. 1. There was no significant change between samples with time;  $p = 0.57$ .

**Table 1** Population pharmacokinetic and model parameters for reduced and total *N*-acetylcysteine (NAC) from 24 male subjects

Model parameter	Value	CV (%) <sup>a</sup>
Total <i>N</i> -acetylcysteine		
CL (whole-body clearance)	$\theta_1 - (\text{exer} \times 0.038)^b$	10.7
Q2 (intercompartmental clearance)	$\theta_2$	23.9
Q3 (intercompartmental clearance)	$\theta_3$	10.9
V1 (central distribution volume)	$\theta_4$	31.2
V2 (peripheral distribution volume)	$\theta_5$	10.3
V3 (peripheral distribution volume)	$\theta_6$	21.8
Parameter estimates	Value	SE
$\theta_1$	$164 \text{ l}^{-1} \text{ kg}^{-1} \text{ h}^{-1}$	0.018
$\theta_2$	$0.123 \text{ l}^{-1} \text{ kg}^{-1} \text{ h}^{-1}$	0.014
$\theta_3$	$0.43 \text{ l}^{-1} \text{ kg}^{-1} \text{ h}^{-1}$	0.013
$\theta_4$	$0.037 \text{ l}^{-1} \text{ kg}^{-1}$	0.009
$\theta_5$	$0.21 \text{ l}^{-1} \text{ kg}^{-1}$	0.022
$\theta_6$	$0.035 \text{ l}^{-1} \text{ kg}^{-1}$	0.008
$\theta_7$	$-0.038 \text{ l}^{-1} \text{ kg}^{-1} \text{ h}^{-1}$	0.014
Within individual variability	19.6%	
MWR	4.18%	
MAWR	24.5%	
Model parameter	Value	CV (%)
Reduced <i>N</i> -acetylcysteine		
CL (whole-body clearance)	$\theta_1 - (\text{exer} \times 0.14)$	12.3
Q2 (intercompartmental clearance)	$\theta_2$	46.2
Q3 (intercompartmental clearance)	$\theta_3$	19.5
V1 (central distribution volume)	$\theta_4$	44.0
V2 (peripheral distribution volume)	$\theta_5$	23.8
V3 (peripheral distribution volume)	$\theta_6$	16.8
Parameter estimates	Value	SE
$\theta_1$	$0.58 \text{ l}^{-1} \text{ kg}^{-1} \text{ h}^{-1}$	0.07
$\theta_2$	$1.01 \text{ l}^{-1} \text{ kg}^{-1} \text{ h}^{-1}$	0.46
$\theta_3$	$0.063 \text{ l}^{-1} \text{ kg}^{-1} \text{ h}^{-1}$	0.012
$\theta_4$	$0.064 \text{ l}^{-1} \text{ kg}^{-1}$	0.028
$\theta_5$	$0.125 \text{ l}^{-1} \text{ kg}^{-1}$	0.03
$\theta_6$	$0.14 \text{ l}^{-1} \text{ kg}^{-1}$	0.023
$\theta_7$	$-0.14 \text{ l}^{-1} \text{ kg}^{-1} \text{ h}^{-1}$	0.039
Within individual variability	24.3%	
MWR	-8.1%	
MAWR	27.5%	

<sup>a</sup>Within and between individual variability are given as CV%

<sup>b</sup>Exer is an indicator variable; exer = 1 during exercise and exer = 0 otherwise

### Pharmacokinetic analysis

Both total and reduced NAC concentration–time data were best fitted to three-compartment open models with covariate effects. The final parameter estimates obtained from the NONMEM analysis are shown in Table 1. The only significantly influential covariate in either model was exercise. This was modelled as an additive effect since including exercise in the model as a graded variable did not improve the fit, as determined by the objective function. For clearance, exercise was modelled as:

$$\text{TVCL} = \theta_1 + \theta_5 \times \text{exer.} \quad (5)$$

Where TVCL is the population estimate for clearance,  $\theta_1$  and  $\theta_5$  are the parameters to be estimated, and exer is an indicator variable that is 1 during exercise and zero otherwise.

Clearance (CL) of NACT was reduced by 23.2% (mean 6.1–40.3; 95% confidence limits) during exercise. The clearance of NACR was also reduced during exercise by 24.7% (mean 11.1–38.2). The small influence of exer-

cise on the central volume of distribution ( $V_1$ ) for NACT was not significant, with the confidence limits including zero. The other covariates  $\dot{V}O_2$ ,  $\dot{V}O_{2\text{peak}}$ , and age did not significantly influence the PK parameter estimates.

Considerable improvements in both MWR and MAWR were seen with the three compartment model as compared with the simpler model. For NACR, MWR was reduced from 17.7 to  $-8.1\%$ , and MAWR was reduced from 47.6 to 27.5%. Improvements of similar magnitude were seen with NACT.

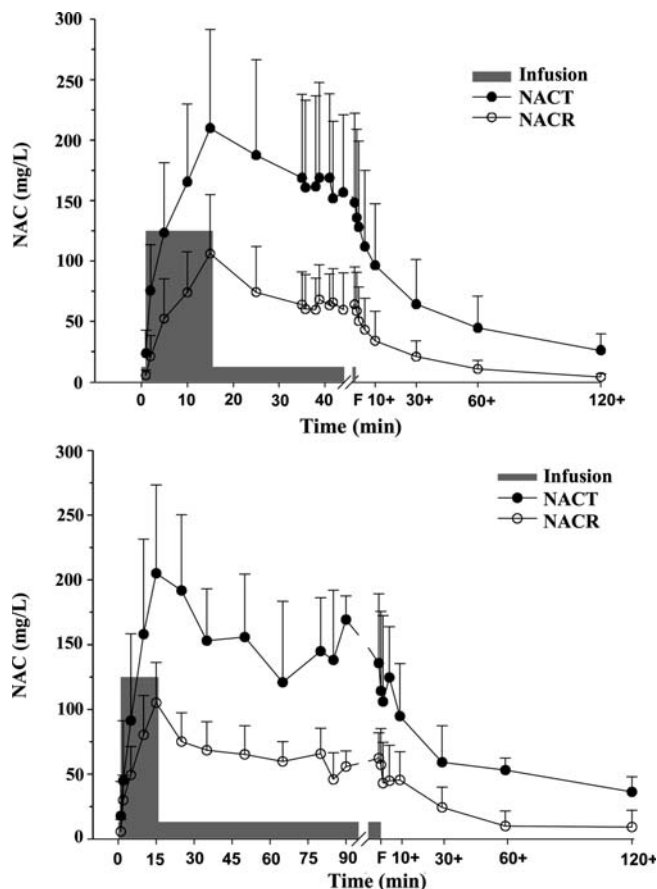
#### Drug concentrations and time course

In all subjects, peak plasma concentrations were seen at the termination of the initial loading infusion ( $125 \text{ mg kg}^{-1} \text{ h}^{-1}$  for 15 min). The mean peak concentration of NACR was  $105.1 (31.1) \text{ mg l}^{-1}$  [mean (SD)] and for NACT was  $205.1 (68.3) \text{ mg l}^{-1}$ . The highest concentrations seen in all subjects were 181.9 and  $310.5 \text{ mg l}^{-1}$ , respectively. Following the first infusion, plasma concentrations fell rapidly and reached a relatively stable plateau within 15–20 min during the second infusion ( $25 \text{ mg kg}^{-1} \text{ h}^{-1}$ ), maintaining concentrations of more than  $50 \text{ mg l}^{-1}$  for NACR and more than  $150 \text{ mg l}^{-1}$  for NACT (Fig. 2). During the exercise phase, there was a transient rise in plasma NAC concentration in all subjects. Following the termination of the longer drug infusion, plasma concentrations declined approximately exponentially.

#### Discussion

Our original intention to evaluate an infusion protocol for NAC suitable for use in high-intensity and/or prolonged lower-intensity exercise has been achieved with the additional benefit of deriving PK data for the intra-infusion and immediate postinfusion periods. The data originally chosen for use as the basis for designing an infusion protocol was taken from a study of the pharmacokinetics of NACT in the treatment of paracetamol overdose [7]. Since the drug was infused in the reduced form and measured as both NACR and NACT, it was possible to derive PK data for both forms of the drug. NACT is not a single species but a mixture of oxidized and reduced dimers and monomers. For the PK analysis, we considered the NACT as an entity since NACT was determined after back-reduction of the oxidized species [2].

For NACR, we estimated CL to be  $0.58 \text{ l kg}^{-1} \text{ h}^{-1}$  compared to  $0.84 \text{ l kg}^{-1} \text{ h}^{-1}$  [6]. The three-compartment fit gave half-lives of 1.3, 15.9, and 101.6 min compared to 8.74 and 117 min [6]. The value we found for NACT clearance was  $0.164 \text{ l kg}^{-1} \text{ h}^{-1}$  compared to the previously reported values of 0.11 [6] and 0.191 [7]. The three half-lives for NACT were 1.5, 11.6, and 132 min. The latter two half-lives are consistent with previous results [6]. We failed to find a longer terminal half-life for either species, and we consider that this is probably due to our short postinfusion observation period. It is possible that



**Fig. 2** Plasma concentrations of *N*-acetylcysteine (NAC): reduced (NACR) and total (NACT). The upper plot shows the data from study 1 (high intensity,  $N=8$ ) and the lower from studies 2 and 3 (prolonged,  $N=16$ ) exercise. Each point is the mean with SD. The NAC infusions are shown as hatched areas proportional to the infusion rate. *F* is the time of fatigue. Duration of exercise (not shown) was 11.4 (SD 1.5) min for study 1 and 62.5 (SD 8.1) min for studies 2 and 3

the inclusion of more data from the elimination phase postinfusion could have resulted in defining this parameter. However, our aim was to obtain data for use during drug infusion, and as with any model, it is strictly applicable only to the subjects and circumstances under which the data were obtained. The finding that both species have very short initial half-lives of 1–2 min may indicate rapid uptake or redistribution into well-perfused tissues, including lung.

We infused a mean total amount of **4,473 mg** in our subjects and obtained mean peak concentrations of  $105.1 \text{ mg l}^{-1}$  for NACR and **205.1  $\text{mg l}^{-1}$**  for NACT. This compares with a 200-mg intravenous bolus achieving corresponding concentrations of 12.2 and  $19.7 \text{ mg l}^{-1}$  [6]. Despite these marked differences in mode of administration and dose, the comparable values obtained for the PK parameters are consistent with the pharmacokinetics of NAC being essentially linear over a wide range and that comparable plasma concentrations within this range should be achievable by scaling dosage and infusion rates between subjects.

**Table 2** Adverse effects to both *N*-acetylcysteine (NAC) and saline in 24 male subjects. Effects have been graded into mild, moderate, or severe. Mild and moderate did not require interruption of the protocol or treatment. There were no severe adverse effects seen that would have required intervention or stopping the infusion. Each subject received NAC and saline on different occasions

Type	Type of reaction and severity							
	NAC				Saline control			
	None	Mild	Moderate	Severe	None	Mild	Moderate	Severe
Incidence of adverse reactions								
Nausea/vomiting	22	1	1	0	23	1	0	0
Local erythema	14	10	0	0	17	7	0	0
Local edema	10	14	0	0	24	0	0	0
Flushing	20	4	0	0	23	1	0	0
Rash	22	2	0	0	22	2	0	0
Coughing	23	1	0	0	23	1	0	0
Bronchospasm	24	0	0	0	24	0	0	0

### Changes during exercise

The clearances of both NACR and NACT were reduced during exercise. The mechanism, although unclear, may be related at least in part to changes in hepatic blood flow. Hepatosplanchnic blood flow measured using indocyanine green is reduced by more than 50% from resting values during cycling exercise of greater intensity than 70%  $\dot{V}O_{2\max}$  and under conditions comparable to this study [14, 15]. We found the clearance of both NACR and NACT to be reduced by about 20% by exercise at 70 and 90%  $\dot{V}O_{2\text{peak}}$ , which is less than the expected reduction in hepatic blood flow. Approximately 70% of NAC clearance is nonrenal [5], and if most of this is hepatic, an estimate of hepatic extraction ratio can be made. In our average 78-kg subject, assuming hepatic plasma flow of 880 ml min<sup>-1</sup> at rest, extraction ratios of 0.6 and 0.17 can be estimated for NACR and NACT, respectively. The latter value is consistent with the estimate of 0.26 [5] based on a smaller estimate of hepatic plasma flow. During vigorous exercise, the reduction of hepatic blood flow would be expected to have a greater effect on the more highly cleared NACR than on NACT [11]. The relatively small reduction is consistent with an increase in extrahepatic clearance of NACR during exercise. We have recently examined the effects of NAC infusion on skeletal muscle NAC and glutathione status during exercise [4]. Our observations support the intramuscular action of NAC as evidenced by both an increase in muscle NAC content and elevated total and reduced muscle glutathione during exercise. We were unable to evaluate the contribution of these effects to the whole-body clearance of NAC.

### Infusion protocol

The intention of using the original infusion protocol was to achieve a target concentration by means of a constant-rate loading infusion and to maintain this target by means of a second constant-rate infusion, with NACT being the marker drug. The NAC concentrations were considerably higher than expected during the initial loading infusion. This was due almost certainly to our

overestimating the value chosen for the initial volume of distribution. The original estimate derived from published data [7] was for a  $V_1$  of 0.283 l kg<sup>-1</sup> and a CL of 0.191 l kg<sup>-1</sup> h<sup>-1</sup>; the final PK estimates of 0.037 and 0.164 l kg<sup>-1</sup> h<sup>-1</sup> indicated that we were close with the clearance estimate but markedly overestimated  $V_1$ , giving NACT concentrations over our intended target of 100 mg l<sup>-1</sup>. For future use of this infusion technique, reducing the rate of the loading infusion should proportionately reduce the target overshoot. A useful outcome of these results is that despite the higher-than-expected NAC concentrations, there were minimal adverse effects of the drug seen (Table 2), as previously reported [2–4], although our data demonstrate that adverse effects graded “mild” were more than twice as common in the NAC phase compared with the control. It is probable that our small cohort of healthy men does not represent an adequate sample from which to extrapolate, particularly given our selection criteria, which were intended to exclude subjects considered to be at increased risk of adverse effects. However, we consider that we achieved a reasonably safe and reproducible method of administering NAC for exercise and related studies. In conclusion, we have estimated the PK of both NACR and NACT at rest and during vigorous exercise. The plasma concentrations achieved were sufficient to cause pharmacological effects [2–4] without causing severe adverse effects due to the drug. We have also shown that exercise has major effects on the pharmacokinetics of NAC.

### References

1. Reid MB, Grubweiser GJ, Stokic DS, Koch SM, Reis AA (1994) *N*-acetylcysteine inhibits muscle fatigue in humans. *J Clin Invest* 94:2468–2474
2. Medved I, Brown MJ, Bjorksten AR, Leppik JA, Sostaric S, McKenna MJ (2003) *N*-acetylcysteine infusion alters blood redox status but not time to fatigue during intense exercise in humans. *J Appl Physiol* 94:1572–1582
3. Medved I, Brown MJ, Bjorksten AR, McKenna MJ (2004) Effects of intravenous *N*-acetylcysteine on time to fatigue and potassium regulation during prolonged cycling exercise. *J Appl Physiol* 96:211–217
4. Medved I, Brown MJ, Bjorksten AR, Murphy KT, Petersen AC, Sostaric S, Gong X, McKenna MJ (2004) *N*-acetylcysteine en-

- hances muscle cysteine and glutathione availability and attenuates fatigue during prolonged exercise in endurance-trained individuals. *J Appl Physiol* 97:1477–1485
5. Borgstrom L, Kagedal B, Paulsen O (1986) Pharmacokinetics of *N*-acetylcysteine in man. *Eur J Clin Pharmacol* 31:217–222
  6. Olsson B, Johansson M, Gabrielsson J, Bolme P (1988) Pharmacokinetics and bioavailability of reduced and oxidized *N*-acetylcysteine. *Eur J Clin Pharmacol* 34:77–82
  7. Prescott LF, Donovan JW, Jarvie DR, Proudfoot AT (1989) The disposition kinetics of intravenous *N*-acetylcysteine in patients with paracetamol overdosage. *Eur J Clin Pharmacol* 37:501–506
  8. Dawson AH, Henry DA, McEwan J (1989) Adverse reactions to *N*-acetylcysteine during treatment for paracetamol poisoning. *Med J Aust* 151:329–331
  9. Mant TG, Tempowski JH, Volans GN, Talbot JC (1984) Adverse reactions to *N*-acetylcysteine and effects of overdose. *Br Med J* 289:217–219
  10. Vaughan DP, Tucker GT (1975) General theory for rapidly establishing steady state drug concentrations using two consecutive constant rate intravenous infusions. *Eur J Clin Pharmacol* 9:235–238
  11. Gibaldi M, Perrier D (1982) *Pharmacokinetics*. 2nd edn. Marcel Dekker, New York
  12. Benet LZ (1972) General treatment of linear mammillary models with elimination from any compartment as used in pharmacokinetics. *J Pharm Sci* 61:536–541
  13. Beal SL, Sheiner LB (1992) *NONMEM user's guide*. NONMEM project group, University of California, San Francisco
  14. Bergeron R, Kjaer M, Simonson L, Bulow J, Skovgaard D, Howlett H, Galbo H (2001) Splanchnic blood flow and hepatic glucose production in exercising humans: role of renin-angiotensin system. *Am J Physiol Regul Integr Comp Physiol* 281:R1854–R1861
  15. Neilsen HB, Clemmesen JO, Skak C, Ott P, Secher NH (2002) Attenuated hepatosplanchnic uptake of lactate during intense exercise in humans. *J Appl Physiol* 92:1677–1683