Study Full Title: A non-inferiority randomised controlled trial of a Shorter Acetylcysteine Regimen for Paracetamol Overdose – the SARPO trial.

Study Short Title: SARPO Trial

Background:

Paracetamol is one of the commonest medications taken in overdose worldwide and is the leading cause of acute liver failure in the United States and Europe[1]. Locally it accounts for 18% of overdose patients presenting to the Princess Alexandra Hospital (PAH) who are managed by the clinical toxicology unit. The antidote acetylcysteine was developed in the 1970's[2] and has decreased both the rates of hepatotoxicity and mortality secondary to paracetamol toxicity. The regimen developed at the time was never subjected to either a randomised controlled trial (RCT) or any dose ranging studies and has remained unchanged until recently[3]. The conventional "3 bag" regimen has been used for decades and is as follows:

1. 150mg/kg NAC over 15-60 minutes
2. 50mg/kg NAC over 4 hours
3. 100mg/kg NAC over 16 hours

From its early clinical use it was recognised that acetylcysteine had adverse effects, which were due to the high initial peak concentration of acetylcysteine attained with the first infusion (150mg/kg) and was anaphylactoid in nature. Since this time a number of studies have been published that have altered the timing of the loading dose and have shown a reduction in adverse effects[3]. One of the largest of these studies undertaken at the PAH in Brisbane, Queensland and the Calvary Mater Newcastle (CMN) hospital in Newcastle New South Wales in 2012-2014, recruited 654 patients and combined the first two infusions (200mg/kg) of the three infusion regimen and administered this over a variable time period of four to nine hours depending on the time of presentation post ingestion[4]. The rate of anaphylactoid reactions was reduced from reported rates of 30-40% to 10% with a 4-hour infusion of 200mg/kg followed by an unchanged 16-hour infusion of 100mg/kg. Although the rate of hepatotoxicity was similar to the traditional regimen, neither this study nor any of the other studies looking at adverse effects were designed or powered to show non-inferiority. This regimen is now used in both toxicology units.

With the development of safer regimens for the administration of acetylcysteine, attention has now been drawn to the original dosing regimen and the evidence behind its dose and duration of administration. The original dosage regimen for acetylcysteine was unpublished but was developed on the following principles[2,5].

1. Patients were presenting with paracetamol hepatotoxicity and were glutathione deficient (70% glutathione depletion required to develop necrosis)[6] and therefore a large loading dose of acetylcysteine as a source of glutathione was required. Glutathione binds to the toxic metabolite of paracetamol *N*-acetyl-*p*-benzoquinone imine (NAPQI). Since these patients had a high rate of morbidity and mortality, high rates of adverse effects were considered acceptable. This contrasts to the current era where nearly all patients with paracetamol overdose present soon after paracetamol ingestion, do not have hepatotoxicity, are not

glutathione deficient and acetylcysteine is administered within eight hours of ingestion.

2. Once the liver had been replenished with glutathione, 6.25mg/kg/hr of acetylcysteine was sufficient based on liver glutathione turnover to maintain glutathione levels[5,7]. In fact this dose "gives a therapeutic excess of acetylcysteine in virtually all cases"[5].

3. The duration of the regimen (20.25 to 21 hours) was set empirically but based on five 4-hour half-lives of a therapeutic dose of paracetamol i.e. acetylcysteine was administered for the duration of time that paracetamol would still be present. This explains the third bag of the acetylcysteine regimen of 100mg/kg/16 hours (6.25mg/kg/hour), which followed the previous two bags given over 4.25 hours. This is despite a half-life of two hours (1.5 to 2.5 hours) when paracetamol is taken therapeutically and 2.9 hours in patients taking a paracetamol overdose who do not have liver damage[8,9]. Longer half-lives are usually only seen in patients with liver damage. More recent evidence of this short half-life comes from PAH clinical toxicology unit patients with paracetamol overdose. In 35 patients ingesting 30g of paracetamol or less with toxic levels, all had paracetamol levels <20mg/L (therapeutic range 10-30mg/L) after 12 hours of acetylcysteine.

The principles of the original acetylcysteine regimen suggest that the management of paracetamol toxicity could be based on ingested dose and half-life of paracetamol (proportional to degree of liver damage) and that one standard regimen should not be used for all patients[5]. Enough acetylcysteine should be given to restore liver glutathione levels and then acetylcysteine should be given while paracetamol is still present. Therefore patients with smaller ingestions and normal livers may require acetylcysteine for less than 20 hours[5]. Patients with liver damage or larger ingestions (prone to prolonged absorption) may require larger doses of acetylcysteine (>6.25mg/kg/hr) for longer than 20 hours since paracetamol will be present for a longer period[5]. Recommendations for larger ingestions have recently been incorporated in the Australia and New Zealand guidelines for paracetamol poisoning[10].

The only study to date that has looked at a shorter regimen was a RCT comparing the traditional regimen with a 12-hour regimen (100mg/kg over 2 hours followed by 200mg/kg over 10 hours)[11]. This study was primarily designed to look at acetylcysteine adverse effects, which were less with the 12-hour regimen. The 12-hour regimen was based on pharmacokinetic modelling that demonstrated a maximum concentration of acetylcysteine (Cmax) that was approximately 20% of the traditional regimen and a acetylcysteine concentration at 20.25 hours being similar to the traditional regimen. The later was subsequently proven to be incorrect[3] when the correct pharmacokinetic model for acetylcysteine was used. It showed the concentration of acetylcysteine to be approximately half the concentration of the traditional regimen at 20.25 hours. Despite this lower acetylcysteine concentration at 20.25 hours, the two regimens had a similar rate of a 50% increase in ALT suggesting similar effectiveness although the study was not sufficiently powered to show non-inferiority.

The authors of the RCT in their discussion state[11]:

"We identified a large proportion of patients with no change in the amount of alanine aminotransferase and with paracetamol concentrations less than 20 mg/L at 12 h. We believe this patient group could be discharged early, if findings of a larger study confirm the absence of inferiority."

Since this publication, there has been discussion in both the United Kingdom[12] and Australia[13] suggesting that low risk patients defined as patients treated within eight hours of ingestion and whose paracetamol concentration is below 20mg/L at the end of a 12-hour acetylcysteine regimen could be safely discharged.

Based on previous research, recent commentary and our understanding of acetylcysteine in paracetamol toxicity, it is proposed to undertake a RCT of two different duration regimens of acetylcysteine. This will provide evidence for the further development of patient tailored regimens of acetylcysteine in paracetamol toxicity.

Aims:

The aims of the study are:

1. To investigate if a shortened 12-hour regimen compared to a 20-hour regimen of acetylcysteine provides the same protection against liver damage from paracetamol overdose in patients taking 30g or less of paracetamol and with an initial paracetamol concentration of less than twice the nomogram line (paracetamol ratio <2 – see below) who commence acetylcysteine within eight hours of ingestion.

2. To assess the adverse reaction rate to acetylcysteine in the first 12 hours.

Hypothesis:

1. That a shortened 12-hour regimen of acetylcysteine in paracetamol overdose will provide the same protection as a 20-hour regimen of acetylcysteine.

2. The rate of acetylcysteine reactions will be similar to previous studies that have utilised an initial 4-hour acetylcysteine infusion of 200mg/kg.

The paracetamol ratio is the first paracetamol concentration taken between four and 16 hours post ingestion divided by the paracetamol concentration on the 150mg/L at four-hour standard nomogram line at the same time point.

Research Plan:

Study Group:

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Angela Chiew	Clinical Toxicologist and Emergency Physician (Prince of Wales Hospital, Sydney, NSW)

Nicole Ryan	Post Doctoral Research Fellow (NHMRC ECF) Clinical Toxicology Research Group University of Newcastle
Geoff Isbister	Clinical Toxicologist and Emergency Physician (Calvary Mater Newcastle, Newcastle, NSW)

Study design and setting:

This will be a multicentre non inferiority per protocol unblinded RCT of a 20 hour versus a 12 hour regimen of acetylcysteine in paracetamol overdose. The study will be undertaken at the PAH, CMN and Prince of Wales hospitals. All three hospitals have established clinical toxicology units, managed by trained clinical toxicologists, which care for all poisoned patients presenting to their respective hospitals. Ethics approval for the three participating sites will be through the Metro South Human Research Ethics Committee located at the PAH. Site specific applications will be through the three respective hospitals involved in the study. Informed consent will be obtained. The study will be registered with the Australian New Zealand Clinical Trials Registry (ANZCTR).

Patient recruitment:

Inclusion criteria:

All patients 16 years and above who fulfil the following criteria:

1. Single immediate release paracetamol overdoses of 30g or less with an initial paracetamol concentration above but less than twice the nomogram line (paracetamol ratio <2)
2. Acetylcysteine can be safely commenced within 8 hours of ingestion.
3. Informed consent can be obtained.

Exclusion criteria

1. Staggered or repeated immediate release paracetamol overdoses.
2. Single, staggered or repeated overdoses of sustained release paracetamol.
3. Repeated supratherapeutic ingestion of paracetamol.
4. Late presentation i.e. >8 hours since ingestion time[a].
5. >30g paracetamol[b] or paracetamol ratio >2.

a. Evidence from published studies of reduced acetylcysteine effectiveness when administered more than 8 hours after paracetamol ingestion[14-16].

b. Recent increasing evidence that ingested paracetamol doses >30g need an increase dose and duration of acetylcysteine[10].

Protocol:

All patients will be commenced on the current 20 hour regimen used at the participating hospitals based on the previous study undertaken at PAH and CMN[4]. That is:

200mg/kg of acetylcysteine in 500mL of 5% glucose over four hours followed by 100mg/kg of acetylcysteine in 1L of 5% glucose over 16 hours (6.25mg/kg/hr).

Once commenced on acetylcysteine and informed consent has been obtained, patients will be randomised to receive either the full 20 hours of acetylcysteine (standard treatment arm) or the first 12 hours (experimental arm) of the 20 hour acetylcysteine regimen. Randomisation can occur at any time up to the point where the patient has received 12 hours of acetylcysteine. Those randomised to receive 12 hours of acetylcysteine only will have their 16 hour infusion of acetylcysteine ceased at eight hours (250mg/kg acetylcysteine) and then be commenced on the equivalent fluid and volume but not acetylcysteine for the remaining eight hours i.e. 500mL of 5% glucose over 8 hours.

Liver function tests will be taken 24 hours post ingestion, which will be approximately two hours prior to the end of the infusion. An additional sample will be taken 12 hours after commencement of acetylcysteine (when the infusion will be ceased in patients randomised to the experimental arm). Patients with acute liver injury defined as ALT >50IU/L and double the admission value[10] at 24 hours post ingestion will have their acetylcysteine continued or recommenced depending on randomisation arm. Patients randomised to the standard arm (20 hours) will continue acetylcysteine at 100mg/kg over 16 hours until the patient is clinically improving, ALT levels are decreasing, the international normalised ratio (INR) is improving and less than 2[10]. Patients randomised to the experimental arm (12 hours) will receive 100mg/kg over 8 hours to catch up and then continue at 100mg/kg over 16 hours. The criteria for ceasing acetylcysteine in the experimental arm will be the same as the standard arm..

Investigations:

1. Paracetamol level and LFT's between four and eight hours post ingestion
2. Paracetamol level and LFT's 12 hours post commencement of acetylcysteine.
3. Paracetamol level, LFT's, INR and miRNA-122 at 24 hours post paracetamol ingestion.

Enrolment, randomisation and blinding:

Emergency department medical staff will be informed and educated on the study and the clinical toxicologists on call will identify suitable patients from the three clinical toxicology units. Enrolment will require contacting the lead investigator (CP) or one of the other investigators (GI or AC) if CP is unavailable. The lead investigator will keep a record of all prospective enrolments. Once contacted and informed consent has been confirmed randomisation will be done by a secure on line website.

Randomisation will be stratified by paracetamol ratio (≤1.5 and >1.5) and also by site. Dose stratification is required so that by chance a similar distribution of overdose amounts is achieved in each acetylcysteine arm. Site stratification will also allow for any differences in the outcome measure analysis by the three hospital laboratories.

Patient's who receive activated charcoal are eligible to be included in the study but they will not be stratified by its use for randomisation. As only a small number of patients currently receive activated charcoal, any effect of charcoal will likely be underpowered, but this will still be examined as a post hoc analysis. Apart from acetylcysteine adverse effects the primary and secondary outcomes measures are objective and laboratory based i.e. liver function tests. Blinding of patient, treating clinician or investigator is not required. However, there will be blinded allocation and once randomisation is done this is recorded online and can't be changed. The acetylcysteine reaction rates are recorded for the first 12 hours only and should be unaffected by treatment arm randomisation and lack of blinding.

Data collection:

A data collection form will be employed at all three sites. The form will include basic demographics, acetylcysteine allocation arm (12 or 20 hours), overdose details including dose and ingestion time, baseline and 24 hour post ingestion liver function tests. An acetylcysteine observation table to record adverse effects will also be part of the form.

Study Outcomes:

Primary outcome:

The primary outcome will be a comparison between the standard and the experimental arm of the absolute difference between the alanine aminotransferase (ALT) on admission and 24 hours post ingestion.

Secondary outcomes:

1. Proportion of patients with a 50% increase in ALT over the admission ALT at 24 hours post ingestion[11].

2. Proportion of patients with an ALT >150IU/L and double the admission value (acute liver injury) at 24 hours post ingestion[17].

3. Proportion of patients with an ALT >1000IU/L (hepatotoxicity) at any time post ingestion[17] assuming it did not rise to >1000 if no change after 24 hours.

4. Differences in other biomarkers apart from ALT. Specifically miRNA-122 and INR at 24 hours post ingestion.

5. Proportion of patients with systemic hypersensitivity reactions in the first 12 hours of treatment with acetylcysteine.

6. Proportion of patients with gastrointestinal adverse effects in the first 12 hours of treatment with acetylcysteine.

Secondary outcomes 5 and 6 are included to enlarge our data set on patients receiving 200mg/kg acetylcysteine over fours hours. Both arms of this study will receive 200mg/kg over four hours.

Post-hoc sub group analysis (a priori)

1. Proportion of patients with an ALT > 50IU/L and double the admission value (acute liver injury) OR paracetamol concentration > 20mg/L after 12 hours of acetylcysteine.

Sample size calculation and statistical analysis:

There are two methods for setting the equivalence boundary or minimally significant effect size for sample size calculation in a non-inferiority trial – clinical and statistical[18,19]. A clinical equivalence boundary is when the researchers and/or a group of clinicians choose the smallest or minimum clinically important difference that they think is important. There are no validated rules for calculating this margin and hence is prone to an arbitrary decision, which is open to differences in interpretation and possible disagreement. A statistical equivalence boundary is based on previous data of the existing treatment effect that the new alternative treatment is to be compared. The non-inferiority margin should be no more than half of the lower limit of the 95% confidence interval of the standard treatment (20 hours acetylcysteine) effect[18].

ALT data from 121 paracetamol overdoses (single ingestion, 30g or less and treated with 20 hour acetylcysteine regimen within 8 hours of ingestion) from the three participating hospitals has been collected. The mean difference between admission and 24 hour ALT is 0.2IU/L with a standard deviation of 10.9 and 95% confidence interval of -21.2 to 21.6 IU/L. Half the lower limit of the 95% confidence interval is 10.7 hence 10 or less should be chosen as the non inferiority margin. A margin of 5 has been chosen which is also satisfactory for a clinical significance boundary. Therefore a mean difference in ALT (between baseline and 24 hours post ingestion) of -4.8 to 5.2 (0.2 +/- 5) in the new treatment arm (12 hour acetylcysteine regimen) would be considered as a non-inferior treatment of paracetamol toxicity.

A non-inferiority study aims to show that one treatment is not significantly worse than another treatment. Therefore this is a one sided test and the significance or alpha level is set at 0.025. With a power of 90% (higher power to minimize the risk of a non-inferior treatment being missed due to chance) and a standard deviation of 10.9 with a non-inferiority limit of 5, the total sample size required is 200 or 100 in each arm[20]. Allowing for a 10% margin for failure to adhere to the study protocol, we aim to recruit 220 patients in total.

The continuous primary outcome of ALT difference between the two-acetylcysteine regimens will be analysed per protocol by student's t-test or non-parametric equivalent depending on the distribution of the data. Secondary outcomes will be analysed using Chi-square or Fisher's exact test, whichever is appropriate.

Study Monitoring

All patients involved in the study will have their liver enzymes closely monitored by the lead investigator and/or the respective site investigator (CP-PAH, GI - CMN and AC - POW) and a Data and Safety Monitoring Board. The committee chair will be Professor Andrew Dawson a clinical pharmacologist and toxicologist from the Royal Prince Alfred (RPA) Hospital (Sydney). There will be three other members. Professor Peter Pillans, clinical pharmacologist from PAH, Professor Tony Brown, emergency physician from Royal Brisbane Woman's Hospital and Dr Joel Oedema, clinical pharmacologist and general physician from Redlands Hospital. The board will meet six monthly or more urgently if required. A report will be supplied to the ethics committee after each review. In the event that the research team and the data monitoring committee feel that the rate of hepatotoxicity in the experimental arm is not consistent with a non-inferior treatment the study may be ceased.

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