STATISTICAL ANALYSIS PLAN (Version 1.04)

Protocol Title:	A randomised, placebo-controlled trial of oxpentifylline on erythropoietin resistance in patients with erythropoietin-resistant anaemia (Version 2.1).
Short title:	HERO: Handling Erythropoietin Resistance with Oxpentifylline.
Protocol Date:	14 March 2011
Trial Registration:	Australian New Zealand Clinical Trials Registry number ACTRN12608000199314

Australasian Kidney Trials Network
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DOCUMENT HISTORY

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APPROVALS

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PREFACE

This Statistical Analysis Plan (SAP) describes the planned analyses and reporting for The Australasian Kidney Trials Network (AKTN) protocol 06.03, A randomised, placebo-controlled trial of oxpentifylline on erythropoietin resistance in patients with erythropoietin-resistant anaemia: The HERO Study.

The structure and content of this SAP provide sufficient detail to meet the requirements identified by the International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH): Guidance on Statistical Principles in Clinical Trials.(1) All work planned and reported for this SAP will follow national and international guidelines for statistical practice (2-5).

The planned analyses identified in this SAP will be included in future manuscripts. Exploratory analyses not necessarily identified in this SAP may be performed to support planned analyses. Any post-hoc or unplanned analyses not specified in this SAP will be clearly identified as such in the Final Statistical Report (FSR) and manuscripts for publication.

This SAP was written by a statistician and clinical investigators who were blinded to treatment allocation and treatment-related study results and will remain so until the central database is locked and the final data are extracted for analysis. To ensure blinding, treatment allocations are stored in a separate location accessible only by an un-blinded AKTN statistician.

The following documents were reviewed when preparing this SAP:

- Clinical Research Protocol for AKTN Trial Number 06.03 (6).
- Case report forms (CRFs) for AKTN Trial Number 06.03.
- Data Safety Monitoring Board (DSMB) Terms of Reference for AKTN Trial Number 06.03 (7).
- ICH Guidance on Statistical Principles for Clinical Trials (1).
- ICH Guidance on Structure and Content of Clinical Study Reports (8).

Readers of this SAP are encouraged to read the Clinical Research Protocol for details on the conduct of this study and the operational aspects of clinical assessments and timing for completing a patient in this study.

ABBREVIATIONS

ABBREVIATION	DEFINITION
ADR	Adverse Drug Reaction
AKTN	Australasian Kidney Trials Network
CKD	Chronic Kidney Disease
DSMB	Data and Safety Monitoring Board
DPO	Darbepoetin
EPO	Erythropoietin
ESA	Erythropoiesis Stimulating Agent
FSR	Final Statistical Report
ICH	International Conference on Harmonisation
ITT	Intention-To-Treat
LOCF	Last Observation Carried Forward
SAE	Serious Adverse Event
SAP	Statistical Analysis Plan
TMC	Trial Management Committee

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1. INTRODUCTION

1.1 Background

The development of erythropoiesis stimulating agents (ESA), such as recombinant human erythropoietin (EPO) and darbepoetin alpha (DPO), has resulted in substantial health benefits for patients with end-stage renal failure, including improved quality of life, reduced blood transfusion requirements, decreased left ventricular mass, diminished sleep disturbance and enhanced exercise capacity (9, 10). Unfortunately, a considerable proportion of such patients exhibit a suboptimal haematologic response to ESA, which in most cases is due to inadequate iron supply to the erythron (11). Other known causes of ESA-resistance include infection, neoplasia, severe hyperparathyroidism, aluminium intoxication, vitamin B_{12} deficiency, folate deficiency, inadequate dialysis, myelosuppressive agents, haemoglobinopathies, myelodysplasia and antibody-mediated pure red cell aplasia (12). However, even after exclusion of these conditions, a significant minority (approximately 10%) of patients exhibit ESA-resistant anaemia and have been shown to have greatly increased morbidity and mortality (12). Inhibition of erythropoiesis by cytokines, such as tumour necrosis factor-α (TNF-α) and interferon-γ (IFN-γ) may play an important role in these patients (13).

Although there is no currently effective treatment for patients with ESA-resistant anaemia, oxpentifylline (pentoxifylline) may represent a promising novel therapeutic strategy. Oxpentifylline has been used for more than twenty years in the treatment of peripheral and cerebral vascular diseases because of its potent haemorrheological properties, which include preservation of erythrocyte water and cation content (14, 15). The drug has subsequently been found to exhibit important anti-inflammatory properties, including anti-apoptotic, anti-oxidant, anti-IL6, anti-TNF-α and anti-IFN-γ actions (16-22), which may in turn decrease hepcidin production (23-26). Hepcidin is a negative regulator of Fe absorption by the intestine and Fe release from macrophages and hepatic stores. Reduction in hepcidin increases iron release from macrophages in the bone marrow resulting in improved availability of iron for erythropoiesis (27). These actions appear to be mediated via inhibition of phosphodiesterase (28). Two small, prospective, non-randomized studies have demonstrated that oxpentifylline may significantly improve haemoglobin levels in chronic kidney disease patients with ESA-resistant anaemia (vide infra) (29, 30).

These studies suggest that oxpentifylline may represent a significant advance in the treatment of ESA-resistant anaemia in chronic kidney disease (CKD), but they are limited by their lack of adequate controls and the associated potential for selection, observer and co-intervention biases. A prospective, randomized, double-blind, placebo-controlled trial is required to definitively test the hypothesis that oxpentifylline corrects ESA-resistant anaemia in patients with CKD.

1.2 Study synopsis

The HERO trial is a multi-centre, prospective, randomised, placebo-controlled trial of the effect of oxpentifylline on erythropoietin resistance in patients with erythropoietin-resistant anaemia. Patients were randomised 1:1 to one of two arms:

- The experimental intervention is oxpentifylline (1 x 400 mg tablet daily; Trental®, Sanofi-Aventis, Sydney, Australia). Oxpentifylline is registered in Australia for

treatment of intermittent claudication on the basis of chronic arterial occlusive disease of the limbs. The standard dose is 400 mg three times daily. The dose of oxpentifylline selected for this study (400 mg daily) is lower than the standard dose because the drug may accumulate in renal failure. Moreover, this dose has been shown to be efficacious without significant side effects in 23 end-stage renal failure patients studied by Cooper et al (29) and Navarro et al (30)

- The control intervention is a placebo (1 tablet daily per os).

The planned sample size was 60 patients. To allow for anticipated rates of drop-out and non-compliance, the recruitment target was 110 patients. The HERO trial began recruiting patients from renal units throughout Australia on 22 June 2009. The trial did not reach its recruitment target and, for reasons of infeasibility, stopped recruiting on 24 August 2012. In total, 53 patients were recruited from 14 centres. The final follow-up visit was conducted in December 2012. The central database is expected to be ready for analysis by June 2013.

1.3 Sub-studies

There are three sub-studies associated with the main HERO Study. These are the hepcidin, oxidative stress, and pharmacokinetic sub-studies. The sub-studies are included in the section of this document on study objectives and study outcome variables but details of the statistical analyses are not described in this SAP.

2. STUDY OBJECTIVES

2.1 Primary objective

The primary objective is to determine whether oxpentifylline at a dose of 400 mg daily results in significant improvement in erythropoietin resistance index (ERI) at 4 months compared with placebo therapy.

2.2 Secondary objectives

Secondary objectives are to determine whether, when compared with placebo, oxpentifylline at a dose of 400 mg daily over four months results in:

1) a reduction to the dosage of erythropoietic stimulatory agent (ESA: either erythropoietin or darbepoetin)
2) an increase in haemoglobin concentration
3) increases in ferritin and transferrin saturation
4) a reduction in blood transfusion requirements
5) an acceptable safety profile for treating ESA-resistant anaemia
6) a cost-effective intervention in terms of its potential to improve quality of life for CKD patients and reduce hospitalisations.

2.3 Tertiary (exploratory) objectives

Exploratory objectives include sub-group comparisons and three sub-studies. C-reactive protein (CRP) levels at baseline will be used to form two subgroups. Patients will be classified as having significantly elevated baseline CRP levels versus not, as defined by local laboratory reference ranges. The effect of oxpentifylline on the primary outcome and key secondary outcomes (ESA dosage, haemoglobin concentration, ferritin, transferrin saturation, blood transfusion requirements) will be determined for the CRP sub-groups.

The first of the three sub-studies will determine whether oxpentifylline at a dose of 400mg daily results in a reduction in serum hepcidin. The second sub-study will determine whether oxpentifylline at a dose of 400mg daily results in a reduction in the biomarkers of inflammation and oxidative stress. The third sub-study will perform pharmacokinetic analyses.

3. STUDY DESIGN ISSUES

3.1 Overview

The study is a prospective, double-blind, randomised, placebo-controlled phase-three trial.

3.2 Study population

The population of interest is CKD patients from renal units (outpatients and dialysis) throughout Australia who have ESA-resistant anaemia.

3.2.1 Inclusion criteria

Patients were eligible for inclusion in the study if <u>all</u> the following criteria were met:

1) Stage 4 or 5 chronic kidney disease (on dialysis or estimated GFR < 30 ml/min/1.73 m^2)
2) Haemoglobin concentration ≤ 120 g/L
3) ERI ≥ 1.0 IU/kg/week/gHb for erythropoietin (EPO) treated patients and ≥ 0.005 µg/kg/week/g Hb for darbepoetin (DPO) treated patients
4) Stable EPO/DPO dosage for ≥ 8 weeks
5) 18 years or over
6) Able to give informed consent

3.2.2 Exclusion criteria

Patients were excluded from the study if <u>any one</u> of the following conditions was met:

1) Patients with a history of psychological illness or condition which interferes with their ability to understand or comply with the requirements of the study
2) Pregnancy or breast-feeding
3) Known hypersensitivity to, or intolerance of, oxpentifylline or other methylxanthines such as caffeine, theophylline or theobromine

4) History of ***major*** gastrointestinal bleeding or ***any*** gastrointestinal bleeding in the past 12 weeks
5) Absolute or functional iron deficiency (ferritin < 100 µg/L and/or transferrin saturation < 20%)
6) Vitamin B12 or folate deficiency
7) Parathyroid hormone level > 100 pmol/L
8) Serum aluminium > 2 µmol/L
9) Urea reduction ratio < 65% or single pool Kt/V < 1.0 (haemodialysis patients) or total weekly Kt/V < 1.7 (peritoneal dialysis patients)
10) Presence of systemic haematological disease (including antibody-mediated pure red cell aplasia) or known haemoglobinopathy
11) Active haemolysis
12) Any surgery within the last 6 weeks
13) Infection within 6 weeks of the last dose of antibiotic, acute myocardial infarction or malignancy within the last 6 weeks
14) Melatonin treatment, androgen therapy or blood transfusion within the previous 4 weeks
15) Vitamin C therapy at a dose greater than 1000 mg/day or at a dose which has changed within the last 12 weeks
16) Haemorrhagic stroke or severe haemorrhage within the last 12 weeks
17) Current immunosuppressant use, or immunosuppression during previous 4 weeks

3.3. Sample size

The study was designed to have 90% statistical power to detect a clinically significant difference in ERI of 0.6 after four months of treatment, assuming a population standard deviation of 0.7 IU/kg/week/gHb and an alpha of 0.05. The difference of 0.6 is 20% of an assumed ERI of 3.1 for the placebo group. Thirty patients per group would be needed to detect a difference of 0.6 given the above assumptions. To allow for a 5% drop-out rate and a 20% non-compliance rate, the recruitment target was 110 patients (55 in each group). The assumed ERI of 3.1 for the placebo group is based on an audit of haemodialysis and peritoneal dialysis patients at Princess Alexandra Hospital, Central Northern Adelaide and Renal Transplant Service and the published literature (29, 30). The sample size calculations described here were first described in a protocol amendment after a change to the primary outcome, the latter being haemoglobin concentration in earlier versions of the protocol and its publication (31). The reasons for the change are described in Section 5.3 of this document.

3.4 Treatment allocation

Patients were randomised to one of the two treatment groups in equal proportion within seven days of their screening visit and after signing a consent form. Randomisation was conducted using a web-based randomisation system. The system randomised patients by an adaptive allocation algorithm designed to minimise imbalance in treatment groups across three variables: study site (anticipated to be up to 18), CKD disease stage (4 or 5), and ESA class (Erythropoietin α/β or Darbepoetin). ESA class was added to the randomisation scheme after a protocol amendment; 16 participants were randomised before this amendment.

3.5 Treatment blinding and allocation concealment

Investigators and patients were blinded to treatment assignment. Microbiology staff in local laboratories who performed outcome assessments were blinded to the patient's assigned treatment. To ensure concealment of treatment allocation, randomisation was performed using web-based access to a central database provided through the Australasian Kidney Trials Network (AKTN).

3.6 Schedule of assessments

Test	Timing of assessment					
	Screening	Baseline	1 month	2 month	3 month	4 month
Demographics & medical history	✓					
Pregnancy test (females of child bearing potential)	✓					
Haptoglobin test	✓					
Full physical examination	✓			✓		✓
Heart rate & blood pressure	✓	✓		✓		✓
Adverse drug reaction			✓	✓	✓	✓
Serious adverse event			✓	✓	✓	✓
Medication dispensed		✓	✓	✓	✓	
Study medication compliance			✓	✓	✓	✓
Concomitant medications	✓			✓		✓
Full blood count (incl. reticulocyte count)	✓	✓	✓	✓	✓	✓
Chemistry profile		✓	✓	✓	✓	✓
Iron studies* (incl. TSAT, ferritin)	✓	✓		✓		✓
C-reactive protein		✓				✓
Short-form 36	✓					✓
Hepcidin blood samples		✓				✓
Inflammation & oxidative stress blood samples		✓				✓
Pharmacokinetic blood samples		✓	✓	✓	✓	✓

* No oral or IV iron may be administered for 7 days prior to iron studies being conducted

All assessments were to be conducted before a patient commenced their dialysis session. For haemodialysis patients, assessments were to be conducted after their "short break".

4. STUDY OUTCOME VARIABLES

4.1 Primary outcome

The primary outcome is erythropoietin resistance index (ERI) measured at the end of the four month study period. ERI is calculated as the dose of ESA divided by haemoglobin level (gHb), where erythropoietin dose is measured in IU/kg body weight/week and darbepoetin dose is measured in μg/kg body weight/week. Section 6.7 gives more information on the primary outcome.

4.2 Secondary outcomes

Secondary outcome variables are:

1) dosage of erythropoietic stimulatory agent (either erythropoietin or darbepoetin) at the end of the 4 month study period
2) haemoglobin concentration at the end of the 4 month study period
3) ferritin and transferrin saturation at the end of the four month study period
4) rate of blood transfusion requirements during the 4 month study period
5) rate of adverse drug reactions (ADRs) during the 4 month study period
6) rate of serious adverse events (SAEs) during the 4 month study period
7) Cost-effectiveness of oxpentifylline during the 4 month study period

4.3 Tertiary outcomes

Tertiary outcome variables are:

1) change in hepcidin concentrations from baseline to the four month study period
2) change in levels of biomarkers of inflammation and oxidative stress from baseline to the four month study period
3) pharmacokinetics of oxpentifylline

4.4 Safety outcomes

Aggregate ADR and SAE rates are included among the secondary outcomes. The following are additional safety outcome variables:

1) death due to any cause
2) any life-threatening event
3) any initial or prolonged inpatient hospitalisation
4) any persistent or significant disability/incapacity
5) any important medical event
6) any congenital abnormality/birth defect
7) SAE body system
8) SAE relationship to study medication (none, unlikely, possible, probably)

5. SEQUENCE OF PLANNED ANALYSES

5.1 Interim analyses

SAEs and measures of study conduct and implementation by treatment group have been monitored on a regular basis by a Data and Safety Monitoring Board (DSMB). Only DSMB members and statisticians compiling closed-session reports for DSMB meetings have had access to un-blinded interim data and results. Due to the short observation period and relatively small number of participants, no interim efficacy analyses were planned or conducted.

5.2 Final analyses and reporting

All final planned analyses identified in the protocol and in this SAP will be performed only after the last patient has completed assessments scheduled for the four month study period and the database has been cleaned and locked. A blinded data review was conducted by members of the Trial Management Committee (TMC) prior to locking and archiving the trial database. Treatment allocations will not be un-blinded and no un-blinded analyses will commence until this SAP has been reviewed by the TMC and approved by the Chair of the TMC and the Trial Statistician. Results from the final analyses will be reviewed by the TMC prior to completion of the FSR and subsequent manuscripts. Any post-hoc exploratory analyses performed to provide support for planned analyses but not identified in this SAP will be documented and reported in appendices to the FSR and clearly identified as unplanned analyses in the text of the FSR.

5.3 Changes to statistical information in the trial protocol

The following amendments have been made to statistical information given in the published trial protocol (31):

- The primary outcome was changed from haemoglobin levels to ERI.
- ESA class was not a stratification variable in the original design.
- Ferritin and transferrin saturation were added as secondary outcome variables.

In March 2010, the primary outcome was changed from haemoglobin levels to ERI in order to optimise participant safety due to recently published results from the TREAT trial which suggested that higher haemoglobin levels (and higher ESA dosages) were associated with increased thromboembolic events in CKD patients (32). Ferritin and transferrin saturation were added as secondary outcome variables because improved iron utilisation is a proposed mechanism by which oxpentifylline can affect haemoglobin concentration and ERI.

6. STATISTICAL METHODS

6.1 Analysis principles

All tests of the effect of treatment on outcomes (except analyses based on subsets) will be conducted on an intention-to-treat (ITT) basis or as close as possible to this ideal by using the

'full analysis set' (1). That is, all randomised patients will be analysed in the group to which they were randomised regardless of whether they received the assigned treatment and irrespective of any protocol deviations or violations. Analyses of outcome variables will, however, exclude data from patients who withdraw from study treatment and withdraw consent for use of their data. All primary statistical analyses will be unadjusted and tests of significance will be two-sided. Any departures from ITT will be documented and reported.

6.2 Incomplete follow-up, missing data, and outliers

6.2.1 Incomplete follow-up

For each patient, participation in the study was scheduled to cease at the end of the four month study visit. If a patient experienced severe anaemia (Hb <65 g/L), symptomatic anaemia or the patient's attending physician believed that additional therapy was required (e.g., blood transfusion) before the end of the four month study visit, then the patient was withdrawn from study medication but followed for study outcome assessments where possible.

6.2.2 Missing outcome data

For the primary outcome and numeric secondary outcomes, patients who do not have a measurement taken at the fourth monthly follow-up visit will have their last observation carried forward (LOCF). While LOCF is usually not recommended, only two patients have missing outcome values at the final visit and results are unlikely to be biased by using this method. Sensitivity analyses will be performed to assess this assumption.

6.2.3 Missing baseline covariate data

There are no missing values on baseline variables that are planned for use as covariates in secondary covariate adjusted analyses of treatment effect.

6.2.4 Outliers

Outliers will be identified by examining residual plots. Cases that visually "stand out" will be assessed for possible influence on results and conclusions by comparing results from analyses with and without the outlier(s). Where there are discrepant results from the two analyses, this will be reported and discussed in the FSR and publication manuscripts.

6.3 Data transformations

Blinded screening of continuous outcome variables suggests no data will require transformation; however, any transformations that become necessary will be documented in SAS analysis programs and described in the FSR and publication manuscripts.

6.4 Multicentre study

Fourteen centres have contributed data to this study. Although included as a variable in the randomisation scheme, study centre will not be adjusted for in the main analyses of the

primary and secondary outcomes. Secondary analyses of the primary outcome and numeric secondary outcomes will be adjusted for study centre where possible. Analyses of non-numeric secondary outcomes will not be adjusted for study centre because some centres are too small and won't contribute any events to the pooled data.

6.5 Multiple comparisons and multiplicity

Results from all analyses will be assessed against an alpha of 0.05. There will be no adjustments for multiplicity as there is a pre-defined hierarchy of outcomes and objectives and the influence of individual results on the overall interpretation of the trial will reflect their importance or level within this hierarchy.

6.6 Covariates, subgroups, and subsets

6.6.1 Covariate adjustment

The primary statistical analyses for continuous outcome variables measured at the four month follow-up visit will be adjusted for the baseline measurement of the outcome variable. All other analyses will be unadjusted. The sample is most likely too small to test the robustness of estimates of treatment effect by performing analyses adjusted for other baseline characteristics suspected *a priori* to be associated with the outcome. Any treatment group differences in potentially influential baseline characteristics will be included in the interpretation and discussion of results.

6.6.2 Subgroups

The main analysis for subgroups will be an unadjusted test of the treatment-by-subgroup variable interaction in a statistical model appropriate for the particular outcome (33). These analyses will be performed regardless of the results of the primary tests of the main effect of treatment.

6.6.3 Subsets

Different subsets of patients contributed data for each of the three sub-studies. While patients will be analysed in the group to which they were randomised, these analyses will not conform to the ITT principle as they will not include all randomised patients.

6.7 Derived and computed variables

Erythropoietin dose is measured in international units (IU) per kilogram of body weight per week (IU/kg/week) and darbepoetin dose is measure in micrograms (µg) per kilogram of body weight per week (µg/kg/week). ERI is calculated as the dose divided by a measure of haemoglobin concentration (grams/litre). To obtain comparable measures of ERI, darbepoetin doses were converted to IU by multiplying the dose in µg by 200.

Patient age (in years) at randomisation will be rounded to an integer value after subtracting date of birth from date of randomisation and dividing the result by 365.25 days.

6.8 Data management and analysis software

Most data manipulation, tables, figures, listings and analyses will be documented in SAS programs and performed using SAS® Software version 9.3. Other data preparation and analyses will be documented in Stata programs and performed using Stata® Software version 12.1.

7. STATISTICAL ANALYSES

7.1 Trial profile

All patients who provide informed consent will be accounted for in the FSR. A CONSORT-style flow diagram will illustrate patient progression through the trial from initial screening for eligibility to completion of the final primary outcome assessment. Number (percentage) by treatment group will be given for patients in the ITT population, reasons for study withdrawal, and major protocol deviations and violations.

7.2 Patient characteristics and baseline comparisons

Demographic and other baseline characteristics will be summarised by assigned treatment group. Categorical variables will be summarised by frequencies and percentages. Percentages will be calculated according to the number of patients for whom data are available. Where values are missing, the denominator, which will be less than the number of patients assigned to the treatment group, will be reported either in the body or a footnote in the summary table. Continuous variables will be summarised by mean and standard deviation as well as quartiles.

The following baseline demographic and clinical characteristics will be presented by treatment group: gender, age at randomisation, ethnic origin (Caucasian, ATSI, Maori/Pacific Islander, Asian, Other, Unknown), height, weight, smoking status (never, former, current), chronic kidney disease stage (4, 5), dialysis status (peritoneal dialysis, haemodialysis), failing renal transplant (no, yes), primary cause of end-stage renal failure (diabetes, hypertension, glomerulonephritis, analgesic nephropathy, polycystic kidney disease, interstitial nephritis, obstructive nephropathy, reflux nephropathy, renovascular disease, other, unknown), co-morbid conditions [diabetes mellitus (no, yes), ischaemic heart disease (no, yes), congestive cardiac failure (no, yes), cerebrovascular disease (no, yes), peripheral vascular disease (no, yes)], heart rate, systolic and diastolic blood pressure, abnormal findings at physical examination (no, yes; if yes, specify). Baseline biochemistry (sodium, potassium, chloride, urea, creatinine, albumin, alkaline phosphatase, total bilirubin, gamma GT, aspartate aminotransferase, alanine aminotransferase, lactate dehydrogenase, calcium, phosphate, glucose, bicarbonate), blood investigations (haemoglobin, haematocrit, red cell count, white cell count, mean cell volume, mean cellular haemoglobin, mean cellular haemoglobin concentration, platelets, reticulocyte count) and iron investigations (serum iron, serum ferritin, total iron binding capacity, serum transferrin saturation) will be presented in three separate tables.

7.3 Analysis of the primary outcome

Treatment groups will be compared on ERI at four months using analysis of covariance. The covariate will be baseline ERI. Supplementary analyses will include a complete-case analysis and a linear mixed model analysis of all available monthly ERI data.

7.4 Analysis of secondary outcomes

Treatment groups will be compared on dosage of erythropoietic stimulatory agent (ESA) at four months using analysis of covariance. The covariate will be baseline ESA. Treatment group differences in haemoglobin levels, at four months will be analysed by the same method, adjusting for baseline haemoglobin levels. Ferritin and transferrin saturation at four months will be similarly analysed, adjusting for their respective baseline values.

Blood transfusion requirements, ADRs, and SAEs by treatment group will be analysed using Poisson regression models. If overdispersion is present, a negative binomial model will be used instead. The incident rate ratio and 95% confidence interval from the appropriate model will be reported.

The main economic evaluation will be an incremental cost-effectiveness analysis. This will consist of calculating the cost of delivering the intervention relative to the control, comparing quality of life scores, mortality and number of hospitalisations between groups. These will be combined to produce an incremental cost-effectiveness ratio.

7.5 Analysis of safety outcomes

The relationship of each SAE to study medication will be rated as none, unlikely, possible, or probable. For each SAE, ratings by treatment group will be summarised by frequencies within each rating category. Percentages are unlikely to be meaningful due to the small sample and relatively small overall number of SAEs.

SAEs will be classified according to body system with reference to the Medical Dictionary for Regulatory Activities (MedDRA) central coding dictionary, version 15. Body system by treatment group will be summarised by frequencies within each body system. Percentages are unlikely to be meaningful for the reasons stated above.

7.6 Exploratory analyses

7.6.1 Subgroup analyses

The main analysis for subgroups formed by CRP levels will be an unadjusted test of the treatment-by-subgroup variable interaction in a statistical model appropriate for the particular outcome. Estimates of treatment effects within each sub-group will be reported with their 95% confidence limits.

7.6.2 Analyses for sub-studies

Analyses for each of the three sub-studies are to be described elsewhere.

8. REPORTING CONVENTIONS

8.1 General reporting conventions

All tables, figures and data listings will be presented in portrait orientation, unless landscape orientation suggests that the information is easier to view. Legends will be used for all figures with more than one variable or item displayed. Figure lines should be wide enough to see the line after being copied.

All titles will be centred on a page. The first title line will be the number of the table, figure, or data listing. The second (and if required, third) line will be the description of the table, figure, or data listing. The ICH numbering convention will be used for all TLFs (8).

All tables, figures, and data listings will have the name of the relevant SAS (or Stata) program and a date-time stamp on the bottom of each output. All analysis programs developed for a table, figure, or data listing will be self-contained to facilitate transfer of programs to multiple computing environments. A separate analysis program will be written to produce each table.

8.2 Statistical summary conventions

For tables, sample sizes for each treatment group will be presented as totals in the column header (N=xxx), where appropriate. Sample sizes shown with summary statistics are the number (n) of patients with non-missing values.

Summaries for categorical variables will include only categories that patients had a response in. Percentages corresponding to null categories (cells) will be suppressed. All summaries for continuous variables will include: N, mean, and SD. Other summaries (e.g. median, quartiles, 5%, 95% intervals, CV or %CV) will be used as appropriate. All percentages should be rounded and reported to a single decimal place (xx.x%). If percentages are reported as integers, percentages greater than 0% but <1% will be reported as <1%, whereas percentages greater than 99% but <100% will be reported as >99%. A percentage of 100% will be reported as 100%. No value of 0% should be reported. Any computation of percent that results in 0% is to be reported as a blank. Summaries that include p-values will report the p-value to three decimal places with a leading zero (0.001). P-values <0.001 will be reported as <0.001.

9. TABLES, LISTINGS, AND FIGURES

This section lists the planned tables, listings and figures for the HERO trial. Templates for all planned tables are in a separate document titled HERO SAP Tables, Listings and Figures.

9.1 Planned tables

The following are planned summary tables:

Table 1. Enrolment by study centre
Table 2. Stratification variables by treatment group

Table 3. Withdrawals, protocol deviations and violations by treatment group
Table 4. Demographic and clinical baseline characteristics by treatment group
Table 5. Baseline biochemistry by treatment group
Table 6. Baseline blood investigations by treatment group
Table 7. Baseline iron investigations by treatment group
Table 8. Clinical assessments by treatment group across study visits
Table 9. Primary outcome by treatment group
Table 10. Secondary outcomes by treatment group
Table 11. Change scores (month 4 minus baseline) and confidence intervals for all investigations by treatment group
Table 12. SAEs by treatment group
Table 13. Relationship of SAE to study medication by treatment group
Table 14. SAE body system by treatment group
Table 15. Cost-effectiveness results by treatment group

9.2 Planned listings

The following are planned data and patient listings:

Listing 1. Deaths and life threatening events
Listing 2. Reasons for patients withdrawing from the study
Listing 3. Medication compliance problems requiring patient to come off trial

9.3 Planned figures

The following are planned summary figures:

Figure 1. Flowchart of patient progression through the study
Figure 2. Time trajectories for ERI by treatment group
Figure 3. Time trajectories for dosage of ESA by treatment group
Figure 4. Time trajectories for haemoglobin concentration by treatment group
Figure 5. Time trajectories for ferritin by treatment group
Figure 6. Time trajectories for transferrin saturation by treatment group
Figure 7. Forest plot of effect of treatment on ERI, ESA dosage, haemoglobin concentration, ferritin, and transferrin saturation for CRP subgroups

10. REFERENCES

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2. ASA. Ethical Guidelines for Statistical Practice. Prepared by the Committee on Professional Ethics. 1999.
3. NZSA. Code of Conduct of the New Zealand Statistical Association. 1995.
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5. SSAI. The Statistical Society of Australia Inc. Code of Conduct.
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