APPENDICES

Appendix A: Incident cases of melioidosis and median annual rainfall

This appendix is contained on an autorun CD-ROM. The short animation provides a conceptual aide to explore the relationship between melioidosis and rainfall in the four dimensions of geographical space (latitude/longitude), time and melioidosis cases/rainfall.

CD-ROM contents

melioid *B.*mov (Quicktime format; requires Apple Quicktime Player)
melioid.avi (AVI format; requires Windows Media Player or similar)

Appendix B: Public domain software used in this thesis

CD-ROM contents:

- **Review Manager 4.2**
  (revman42.exe; available at http://www.cc-ims.net/RevMan/)
  RevMan is the Cochrane Collaboration's program for preparing and maintaining Cochrane reviews. RevMan is currently developed at The Nordic Cochrane Centre. The development is overseen by the Information Management System Group and the RevMan Advisory Group.

- **SaTScan 3.0**
  (sts3.0.5.exe; available at http://www.satscan.org/)
  The SaTScan software has been developed to analyze spatial, temporal and space-time count data using the spatial, temporal, or space-time scan statistics. SaTScan was developed by Martin Kulldorff (National Cancer Institute, National Institutes for Health, United States) together with Information Management Systems Inc.

- **Stata modules**
    - **`ralloc`** : Ryan P, Stata module to design randomized controlled trials. Stata Technical Bulletin 54, 1997 (ralloc.ado, ralloc.hlp; available at http://ideas.repec.org/c/boc/bocode/s319901.html)
    - **`ineq`** : Cox, NJ, Stata module to calculate measures of inequality. 1999 (ineq.ado, ineq.hlp; http://ideas.repec.org/p/boc/bocode/s365801.html)
    - **`gam`** and **`gamplot`** : Royston P, Ambler G (1998) Generalized additive models. Stata Technical Bulletin 42: 38-43. (gam.ado, gam.hlp, gamplot.ado, gamplot.hlp; http://ideas.repec.org/p/boc/bocode/s428701.html)

The software was run on a Dell Inspiron 5000 (600MHz, 128 MB RAM) with Windows 98 and a Toshiba Satellite Pro TE2100 (2.0 GHz, 512 MB RAM) using a Windows XP Professional platform. Stata modules were run on Intercooled Stata 7.0 for Windows.

Appendix C: Protocol and consent forms for proposed trial of G-CSF in patients with septic shock due to melioidosis in Ubon Ratchathani

A placebo-controlled trial of granulocyte colony-stimulating factor in ceftazidime-treated patients in severe sepsis due to melioidosis

Collaborating investigators

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Summary

We propose a randomized, blinded, controlled trial of granulocyte colony-stimulating factor (G-CSF) vs placebo in the treatment of melioidosis and severe sepsis.

Background

Melioidosis, an infection caused by the bacterium *Burkholderia pseudomallei*, is a major cause of community acquired septicaemia in north eastern Thailand. Common manifestations are cavitating pneumonia, hepatic and splenic abscesses, and soft tissue and joint infections. (1) Treatment is difficult; despite improvements in the diagnostic procedures and treatment regimens the mortality of severe melioidosis remains unacceptably high. In Thailand, the mortality remains approximately 35% with the currently used antibiotics such as ceftazidime or co-amoxiclav (2) and in Australia, the mortality is approximately 20% (3).

There is a real need for better antibiotic treatment and other treatment modifications to reduce this high mortality. Most of the fatal cases die within 72 hours after admission, even with the best antibiotic treatment, ceftazidime reduced by half the mortality of severe melioidosis (from 70 to 35%) when compared with conventional treatment (doxycycline + chloramphenicol + cotrimoxazole) but it affected outcome only after 48 hours. The mortality in the first 48 hours has not been altered by any treatment regimens (4). A previous study of imipenem vs ceftazidime revealed that although endotoxin release was reduced with imipenem (5), mortality was not affected.

Where most clinical trials of interventions in melioidosis have been in patients with sepsis, much of the mortality occurs in an even more acutely unwell group, those with severe sepsis with markers of end organ dysfunction. Thus, interventions to reduce the mortality from melioidosis might best be targeted at this small group.

G-CSF has been widely used in the treatment of congenital and acquired neutropenias (6, 7). In animal models, prophylactic treatment with G-CSF of non-neutropenic mice reduces sepsis-related mortality (8, 9). G-CSF acts to increase the production of granulocytes and also results in an improvement in neutrophil function. In addition, G-CSF has immunomodulatory effects, suppressing the pro-inflammatory cytokines IL-2 and IFN-γ and stimulating the production of the anti-inflammatory cytokines IL-1ra, sTNF-R and IL-10 (10). Neutrophil function has been shown to be impaired in conditions such as malnutrition (11), diabetes (12), chronic renal failure (13) and hazardous alcohol use, (14) conditions which are more common in our patient population (3, 15, 16).

However, human studies of G-CSF in non-neutropenic hosts have been less impressive; an initial study in severe community acquired pneumonia without sepsis did not improve time to resolution of morbidity, length of hospitalization or mortality, although sub-group analysis suggested that benefits were seen in the group with multilobar pneumonia (17). A follow-up study examining G-CSF in community acquired pneumonia with multilobar involvement did not result in improved clinical outcomes, but subgroup analysis suggested that benefits may be seen in patients with pneumococcal pneumonia (18).
A phase II study of G-CSF in severe community acquired pneumonia with septic shock has showed some clinical benefit, but small numbers (n=18) precluded meaningful analysis (19). A phase III study of G-CSF in pneumonia and severe sepsis (n=701) failed to reveal any benefits associated with G-CSF. (20) It was suggested that delays in administration of G-CSF, due to microbiological confirmation being part of the enrolment criteria, may have contributed to its negative result.

G-CSF was adopted at the Royal Darwin Hospital in December, 1998 in response to the *in vitro* and animal data available at that time. Thus, it was felt that G-CSF may reverse neutrophil dysfunction due to co-morbid conditions in the patient group seen in Australia. This group may differ from the populations studied in the trials mentioned previously in the type of bacterial pathogens, younger age and the presence of co-morbid conditions.
Since that time, uncontrolled use of G-CSF has resulted in a reduction of mortality from septic shock from 73% to 31% (21). More dramatically, of patients with melioidosis with septic shock, mortality has fallen from 95% (20 of 21 patients) to 10% (2 of 21 patients) since the adoption of G-CSF (unpublished data; manuscript attached). However, possible confounding factors include the appointment of an Intensive Care Specialist, the use of physiological dose steroids, early enteral feeding, the earlier use of meropenem in septic patients, and the introduction of management protocols for patients with septic shock. In this audit, it was suggested that a randomized placebo-controlled trial would be required to confirm this apparent benefit.

We acknowledge that there are a number of candidate immunomodulator therapies that may improve mortality from severe melioidosis, including polyclonal immunoglobulin, activated protein C (22) and recombinant tissue factor pathway inhibitors (23). IgM-enriched immunoglobulin did not improve mortality from severe sepsis in a recent trial, (24) and anti-lipopolysaccaride antibodies are of uncertain value in melioidosis. Although polyclonal immunoglobulin is associated

with reduction in sepsis-related mortality (25), its role in melioidosis is as yet untested. Many of these therapies are extremely expensive (activated protein C costs approximately 300,000 baht per patient) and there is debate regarding their cost-benefit ratio even in wealthy countries.

The evidence from Australia suggests that G-CSF may be a promising therapy. Its cost is much less that most immunomodulatory therapies such as activated protein C. It is licenced for use in Thailand and is commonly used in other conditions. If this can be shown to be safe and effective, this will have a large impact for a group with a high mortality.

Objective

We propose a randomized, blinded, control trial to test the use of G-CSF in combination with ceftazidime-based standard antibiotic therapy in the treatment of severe sepsis due to melioidosis.

Place and period

At Sappasitiprasong Hospital, Ubon Ratchatani, North Eastern Thailand between July, 2003 to July 2004.

Materials and Methods

Patients

The study population will include severely unwell patients admitted to the hospital with suspected melioidosis, who need hospitalisation and intravenous antibiotic administration and who have severe sepsis.

Inclusion criteria (all the following criteria must be satisfied)

1. Community acquired severe sepsis (must fulfil criteria for sepsis (1.1) and end-organ perfusion abnormalities (1.2)), and melioidosis is suspected. The criteria for sepsis and shock have widely-used standard criteria for sepsis and shock (26). The markers of end organ perfusion abnormalities are based on an assessment system for organ dysfunction (27, 28) and previous studies of septic shock (22):

1.1 Sepsis: Systemic Inflammatory Response Syndrome: **two or more** of the following, clinically ascribed to infection

Fever: temperature >38^{o}C or <36^{o}C
Tachycardia: heart rate >90 beats/min
Tachypnoea:
Respiratory rate >20 breaths/minute; or
P_aCO_2 <32 mmHg; or
Mechanical ventilation
White cell count >12,000 cells/mL or <4,000 cells/mL or >10% band forms

1.2 Severe sepsis Sepsis (criteria 1.1) with markers of perfusion abnormalities that may include **two or more** of:

Metabolic acidosis : pH<7.3 or base excess <-5 or
Lactate ≥3 mg/L
Respiratory dysfunction: Need for mechanical ventilation or
PaO_2/FiO_2<300
Renal dysfunction:

Oliguria < 500ml/24 hours or
Creatinine >2.0 mg/dL or
Renal replacement therapy
Altered mental status GCS<12 (if not sedated)
Liver dysfunction; bilirubin > 2.0 mg/dL
Thrombocytopenia: platelet count <100,000 cells/mL
Shock: Hypotension for more than 1 hour in absence of other causes of hypotension (eg anaesthesia or antihypertensive medication) despite adequate fluid challenge sufficient to restore circulating blood volume
(Systolic blood pressure <90mmHg; or
Fall of >40mmHg from baseline; or
Requirement for vasopressors/inotropes)
2. Community acquired septicaemia (diagnosis made within less than 72 hours of hospitalization)
3. No known hypersensitivity to G-CSF
4. Age > 14 years
5. Need hospitalisation and intravenous antibiotic administration
6. Willingness to participate in the study and written, informed consent obtained from the patient

2. Exclusion criteria (any one of the following)
1. Patients where the time from diagnosis of severe sepsis exceeds 24 hours.
2. Known haematological malignancy, myelodysplasia or congenital neutropenia
3. Febrile neutropenia (as this group has been shown to benefit from G-CSF (6) and thus would be unethical to give placebo)
4. Pregnant or lactating women
5. Known hypersensitivity to G-CSF
6. Patients not expected to remain in hospital for treatment
7. Known objection to participation in study
8. Patients who have previously been enrolled or who have received G-CSF within the past month
9. Patients with community-acquired sepsis with cultures positive for other organisms

Methods

1. Study Design

This will be a randomized, blinded, controlled trial

Patients will be identified from examination of daily admission logs, ward rounds of medical, respiratory care and intensive care wards, and discussion with treating clinicians. Any patient with suspected melioidosis and severe sepsis will be eligible for enrolment. Patients that do not fulfill these criteria or are specifically excluded from the study will be treated with routine management protocols for melioidosis. Detail of illness and physical examination will be taken for study participants and recorded onto clinical record form attached with this document.

2. Baseline investigations

1. Microbiologic examination including culture from all possible sites: blood, sputum, pus, urine. A direct immunofluorescent antibody test (29) will be performed in any collected clinical specimens.

2. Complete blood count, BUN (urea), creatinine, electrolytes, and liver function tests

3. Other blood tests based on clinical grounds and decisions of treating ward doctors

4. Chest x-ray and other x-ray or ultrasonography based on clinical grounds

The total blood volume taken for baseline investigations will be 10 ml.

3. Treatment

The randomized sequence for the G-CSF trial will be generated by the Computing and Statistics Unit at Menzies School of Health Research using Intercooled Stata 7.0 and will be concealed from all investigators during the study. Block randomization using varying block sizes will be used to minimize any imbalance between the intervention groups.

The study drugs will be labeled with "G-CSF study drug" and numbered then kept in the same labeled, sealed box including the full details of the preparation and dosing for the ward nurse. The study doctors and patients are blinded to the treatment being administered to each patient, and the nurse will be asked not to disclose allocation. The boxes will contain either:

G-CSF: Lenograstim (Recombinant human granulocyte colony stimulating factor or rhG-CSF) (Granocyte®-Chugai Pharmaceutical, Japan) with normal saline (1ml) for dilution

Dose: 263 μg intravenous injection, once daily for 3 days

No dose adjustment in renal impairment

Placebo: Normal 0.9% saline

Dose: 1ml intravenous injection, once daily for 3 days

The patients will be closely observed by the study team for the progression of the disease and any adverse effects of any treatment. Apart from the study medications, patients will receive standard treatment for sepsis and melioidosis, including ventilation, inotropic support, and fluid management. The study antibiotics will be given for at least 10 days or until the patients show the clear improvement in signs and symptoms including clearing of fever for at least 48 hours. Subsequent oral antibiotics will be prescribed for another 20 weeks according to standard treatment guidelines.

4. Other investigations

Investigations specifically performed for this study will be as follows:

Lactate before treatment, then day 2, 4 and 7 (total blood volume 2ml)

Haemoculture at 72 hours and day 7 and then weekly until no *B. pseudomallei* isolated from blood (at least 5ml)

CBC, BUN, creatinine and electrolytes on day 3, 7 and then weekly until the cessation of intravenous drug (at least 10ml)

Culture from other sites at 72 hours and weekly until negative

Cytokines (G-CSF, IL-2 and IL-10) on admission and day 3 (total blood volume 6ml)

White blood count at 24, 48 and 72 hours (total blood volume 1ml)

Neutrophil functions studies on admission and day 3 (total blood volume 40ml)

Total blood volume taken for this study apart from the baseline investigation will be 64 ml.

5. Patient monitoring during study

The patients will be closely monitored by both the clinicians and researchers each day. The physiologic scores will be assessed to adjust for the severity of the disease, such as APACHE II (Acute Physiology and Chronic Health Evaluation) scores (30), SOFA scores (sepsis-related organ failure assessment) (28) at day 1,3,7 and 10. Details of the SOFA score is detailed below:

SOFA	0 points	1 point	2 points	3 points	4 points
Respiratory: PaO2/FiO2	>400	300-400	200-300	100-200	<100
Coagulopathy: Platelets (/ml)	>150	100-150	50-100	20-50	<20
Hepatic: Bilirubin (mg/dl)	<1.2	1.2-1.9	2.0-5.9	6.0-11.9	>12.0
Cardiovascular: Blood pressure (mmHg)	No hypotension	MAP<70	Dopamine<5 or Dobutamine	Dopamine>5, adrenaline<0.1 or noradrenaline <0.1	Dopamine>15, adrenaline >0.1 or noradrenaline >0.1
Neurological: GCS	15	13-14	10-12	6-9	<6
Renal: Creatinine (mg/dl)/ urine output	Cr<1.2	Cr 1.2-1.9	Cr 2.0-3.4	Cr 3.4-4.9 or UO<500mL/d	Cr >5.0 or UO<200mL/d

5. Withdrawal criteria

1. Serious adverse event believed to be attributable to study drug
2. Withdrawal of patient consent
3. Discharge of patient from hospital
4. White cell count exceeding 75,000 cells/mL

Outcome measurement

The primary outcome measures will be in-hospital mortality and 28 day mortality. Secondary outcome measures will include the following:

1. Treatment failure: unfavorable outcome or requirement to change antibiotic regimen according to the following criteria:
Obvious worsening of clinical signs and symptoms after 72 hours of treatment such as lowering of blood pressure, deterioration of shock
B. pseudomallei persistently cultured in blood after 7 days of treatment
Persistent fever and no obvious improvement in any clinical signs and symptoms after 10 days of treatment in the present of other proper management such as surgical drainage of pus or fluid collection.
2. Fever clearance time
3. Adverse drug reactions
4. Sepsis-related organ failure assessment (SOFA) scores (28) at day 1,3,7 and 10
5. Time to resolution of shock
6. Duration of ventilation
7. Duration of hospitalization
8. Neutrophil function before and after treatment

Statistical methods

1. Sample size

Previous work has shown that the mortality of severe sepsis is approximately 65%. We aim to demonstrate a reduction in mortality from 64% to 32% with 80% power. Thus, 88 culture-proven melioidosis cases are needed. Assuming that 80% of patients have culture-confirmed melioidosis, a total enrolment of approximately 110 patients is required for this study. As we believe that severe sepsis is a common endpoint and that the source of infection, whether pneumonia or intra-abdominal suppuration, does not in itself influence mortality, we believe that we can answer the study question with this relatively small sample size.

The sample size calculations, using Intercooled Stata 7.0, are reproduced below:

```
. sampsi 0.64 0.32, a(0.05) p(0.8)

Estimated sample size for two-sample comparison of proportions

Test Ho: p1 = p2, where p1 is the proportion in population 1
                    and p2 is the proportion in population 2
Assumptions:

         alpha =   0.0500  (two-sided)
         power =   0.8000
            p1 =   0.6400
            p2 =   0.3200
         n2/n1 =   1.00

Estimated required sample sizes:

            n1 =       44
            n2 =       44

. di (44+44)/0.8
110
```

2. Statistical methods

Analysis of proportions (such as mortality) will be performed using Fisher's exact test. Continuous data will be assessed for normality; for non-parametric variables, a Mann Whitney U test will be employed to test for the equality of distributions. For the comparison of means, Student's t-test will be used. A multivariate model, controlling for severity of illness (indicated by APACHE 2 score) will be utilized in the mortality analysis. Duration of survival will be analyzed by Kaplan Meier plots and a Cox hazards analysis. Data will be analyzed by Stata 8.0.

6. External monitoring

An external data safety board will perform an interim analysis following enrolment of the 10th patient and the 40th patient.

The interim analysis will assess the following parameters:
Review of eligibility
Design assumptions (rate of enrolment, resource availability)
Baseline comparability of groups
Adherence to treatment protocol
Adverse events

This data will be provided by the study team together with the treatment assignment to allow analysis. The study monitor will not disclose the results of the interim analysis to the study team if the study proceeds.

The stopping rules will be based on the methods of O'Brien and Fleming (1979) where an outcome will be deemed to be significant at a level of 0.0054. Conditional power will be calculated to determine futility by the calculation of three scenarios: that the current trends will continue until the completion of the trial, that the original estimate for efficacy will continue until the end of the trial, and that there is no difference between treatments for the remainder of the trial.

The outcome of the analysis will answer the following questions:
Is there adequate data on which to base a decision?
Should the trial continue?
Should the study protocol be modified?

Ethical considerations

Safety data

Ceftazidime is an antibiotic which is widely used in the treatment of infectious disease. Ceftazidime is the current standard treatment for melioidosis. Renal function is routinely monitored at least every week in the treatment of melioidosis, and the dosage is adjusted according to the creatinine level. Ceftazidime may cause nausea, vomiting, diarrhoea, and phlebitis at injection site. (31)

G-CSF has been used for over 10 years in the treatment of neutropenia, mostly in relation to cancer chemotherapy. Its safety profile is such that it is commonly given to volunteer donors for mobilization of peripheral blood progenitor cells for use in allogeneic transplants. The commonest side effect reported has been mild to moderate bone pain, which is usually controlled with non-opioid analgesia. (6)

There has been a theoretical concern that the use of G-CSF may worsen organ dysfunction, such as ARDS. However, in the three trials of G-CSF in non-neutropenic hosts in pneumonia, enrolling a total of 629 patients in the treatment arms, there was no increase in the incidence of significant adverse events, including ARDS, when compared to placebo (17-20). Other rarely reported adverse effects include rash, cutaneous vasculitis and Sweet's syndrome (32-35).

The procedure to take blood samples and drug injections may cause trauma to the blood vessels, which may lead to ecchymosis or thrombophlebitis. To minimize this

side effect, these procedures will be performed by experienced, well-trained doctors or nurses.
The study team will take the full responsibility for any harm and complications which may be caused by the study procedures, without any cost to the patients.

We will report to the Ethical Committee, Ministry of Public Health immediately if any adverse effects occur due to the study drugs.

Information sheet and Informed Consent
The information concerning the study will be given in detail by a Thai doctor or nurse. The patients will be enrolled after she/he or their relatives agree and sign the consent form.

A copy of the proposed consent form is attached.

The participants or their relatives can withdraw from the study at any time at any reason. After withdrawal, the study drugs will be ceased and ceftazidime will be given at standard doses; the other treatment will not be affected.
The patient information will be kept confidentially by the study team. Only the outcome of the study will be published, not any individual patient information

Ethical clearance
This proposed study will be submitted to The Ethical Clearance from Ministry of Public Health, Royal Government of Thailand, the Human Research Ethics Committee of Territory Health Services and Menzies School of Health Research, Australia for approval.
This proposed study has been approved by the Human Research Ethics Committee of Territory Health Services and Menzies School of Health Research, Australia.

Funding
This study is a collaboration work between The Wellcome Trust-Mahidol University, Oxford University Tropical Medicine Research Programme, Sappasitiprasong Hospital Ubon Ratchatani, Thailand and Menzies School of Health Research, Darwin, Australia.
The study will be funded by the Wellcome Trust of Great Britain, National Health and Medical Research Council of Australia, Royal Australian College of Physicians and Flinders University.
G-CSF used will be partly donated by Merck, Australia and partly purchased with the support of the Australian funding sources.

Budget for the study

Laboratory and clinical expenses	100,000 ฿
Communication	5,000 ฿
Transportation	50,000 ฿
Total expenditure	155,000 ฿

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INFORMATION SHEET

A placebo-controlled trial of granulocyte colony-stimulating factor in ceftazidime-treated patients in severe sepsis due to melioidosis

You/your relative is being asked to join a research study of patients who have severe melioidosis. We would like to see if a medicine, called granulocyte colony stimulating factor ("G-CSF") will help the body fight off this infection. This medicine has been used safely in patients with severe melioidosis in Australia but not in Thailand. However, we do not know if it makes any difference.
In order to decide whether you/your relative wishes to be in this study, you will need to know about the study and the effects of the medicine we are using. This sheet tells you about the study. You can ask the interviewer any questions you might have at any time.

Why we're doing this study

We believe that white blood cells, the cells that the body produces to fight infection, react differently to infection in each person. G-CSF is sometimes used to increase white cell numbers in patients that have had a problem with low numbers, such as after cancer chemotherapy. G-CSF also makes white cells work better in people with normal numbers of white cells, but with a severe infection.
In Australia, patients have been given G-CSF for melioidosis which seemed to result in less patients dying from melioidosis. However, it is difficult to know if this was the result of this medicine, or other changes that occurred at this time. Studies performed elsewhere in the world have not shown any benefit with this medicine, but there are differences between those studies and local conditions. Because of these concerns, G-CSF has not been used in patients with melioidosis other than in Australia.

The best way to find out if G-CSF works is to give G-CSF to some patients and comparing this group with patients who are not given this medicine. In this type of study, called a randomized controlled trial, some patients are given G-CSF and others are not. We will not know which patients are receiving G-CSF until the end of the study where we will be able to see if the G-CSF made any difference to the success of fighting off the infection.

Safety data

Ceftazidime is the current standard treatment for melioidosis; all patients in this study will receive this antibiotic. The dose using in treatment of melioidosis is a high dose, which may affect renal function. In practice the renal function are routinely monitored every week in the treatment of melioidosis, and the dose will be adjusted according to the creatinine level. Ceftazidime may cause nausea, vomiting, diarrhoea, and phlebitis at injection site.
G-CSF has been used for over 10 years in the treatment of neutropenia, mostly in relation to cancer chemotherapy. Its safety profile is such that it is commonly given to volunteer donors for mobilization of peripheral blood progenitor cells for use in allogeneic transplants. The commonest side effect reported has been mild to moderate bone pain, which is usually controlled with simple painkillers. Other possible adverse effect are chest pain, anorexia, fever, headache, rash, cutaneous vasculitis.

What we're asking you to do

In this study, you will be asked a number of questions about your medical history and risk factors for infections. This will take less than 15 minutes and will be completed with the help of your medical team. Some of these questions may be personal. You can choose not to answer any of these questions for any reason; just say so and we will move on to the next one.

You will be asked to give consent for our staff to collect data from you and your medical records that relates to the study. In addition to the routine blood tests that you receive as part of your care here, an additional 64 mL (about 4 tablespoons and 1 teaspoon) of blood will be taken to be stored and tested within the first week of study. In addition, any other body samples that are obtained while you are here may be stored for future testing. Apart from taking extra blood samples and receiving the study medicine, your medical care will not be changed by your being part of the study.

Your information and details will be kept confidential by the study team. If any complications occurred as part of the study the doctors would be responsible and any treatment needed without any cost.

You have the rights to ask at any time. After the participation you can withdraw at any time and for any reason. The study drugs will be ceased and the standard treatment for melioidosis, ceftazidime, will be continued. Any other treatments will not be affected and you will continue to be managed by the treating doctors..

The drug for the study will be provided by the study doctor this includes the whole course of intravenous antibiotic treatment. The oral antibiotic drugs for the maintenance treatment are not included in the study, so this will be the usual cost for you. The result of this study will help us further improve the treatment of melioidosis.

Name of 24 hours contacts in case of any problems or questions:

1. Dr. Wirongrong Chierakul: Melioidosis Laboratory, Sappasitiprasong Hospital. Tel: 045 246112, Mobile phone: 09 1058571
2. Dr. Adul Rajanuwong: Medical Department, Sappasitiprasong Hospital. Tel: 045 244973, Mobile phone: 01 7172703
3. Dr. Allen Cheng: Melioidosis Laboratory, Sappasitiprasong Hospital. Tel: 045 246112

CONSENT FORM

A placebo-controlled trial of granulocyte colony-stimulating factor in ceftazidime-treated patients in severe sepsis due to melioidosis

Date..... HN..... Ubon no.....

Name..... Age.....

Address.....

.....

I understand and have been given an information sheet on this research study and have discussed the study with _____

(name of doctor or nurse)

I am aware of the procedures involved in this study, including:
Administration of ceftazidime with G-CSF or placebo
Blood draws 74ml within first week (4 tablespoons and 1 teaspoon)
These procedures have been explained to me, including indications of risks; any discomfort involved; anticipation of length of time and the frequency with which the procedures will be performed. I have understood and am satisfied with the explanations that I have been given. I have been provided with a written information sheet.

I understand that my involvement in this research project may not be of any direct benefit to me and that I may withdraw my consent at any stage without affecting my rights or the responsibilities of the researchers in any respect.

I have the right to withdraw from the study at any time and for any reason. There will be no effects to the management which I should have.

I understand that the researchers will take the responsibility for any harm or complications which may happen from the study procedures without any cost to me.

My information will be kept confidentially by the researcher. Only the outcome of the study can be presented or published, not my individual information.

I can contact the researchers at any time in case of any questions or problems:
1. Dr. Wirongrong Chierakul: Melioidosis Laboratory, Sappasitiprasong Hospital. Tel: 045 246112, Mobile phone: 09 1058571
2. Dr. Adul Rajanuwong: Medical Department, Sappasitiprasong Hospital. Tel: 045 244973, Mobile phone: 01 7172703
3. Dr. Allen Cheng: Melioidosis Laboratory, Sappasitiprasong Hospital. Tel: 045 246112

I have read the document and hereby agree to participate in this research study.

.....Patient
.....Researcher
.....Witness
.....Witness

I cannot read, but the researcher has read the document for me. I have listened and clearly understand. I hereby agree to participate in this research study.

.....Patient
.....Researcher
.....Witness
.....Witness

In case of age under 20, the guidance or parents must be consented and sign.

.....Guidance/Parents

In case of patients unable to give consent, relatives/parents consent and sign.

.....Patient
.....Researcher
.....Witness
.....Witness

VITA

Allen Cheuk-Seng Cheng was born in Melbourne, Australia in 1970.

He received a Bachelor of Medicine and Surgery at the University of Melbourne (1988-1993) and a Graduate Diploma in Clinical Epidemiology at Monash University (1999-2000). He is a Fellow of the Royal Australasian College of Physicians.

Allen completed his residency at St Vincent's Hospital Melbourne (1994-1998). During this time, he also worked in Dumfries and Galloway Royal Infirmary, Scotland and the Royal Darwin and Katherine Hospitals (1996). He completed advanced training in infectious diseases at the Alfred Hospital Melbourne (1999-2000) and Duke University Medical Center, North Carolina, United States (2000-01).

He has also worked as a consultant to the Medical Officer, Nursing and Allied Health Training Project based at the Port Moresby General Hospital, Papua New Guinea (1998, 1999, 2002). Currently, Allen is a general physician to the Groote Eylandt and Bulman communities in the Northern Territory as well as an Honorary Research Fellow and Deputy Director of Clincal Studies at the Wellcome Trust-Oxford University-Mahidol University Tropical Medicine Research Unit in Ubon Ratchathani, Thailand.

Allen received an Exhibition in Medical Chemistry (University of Melbourne, 1988), the Andrew Brenan Prize in Pathology (St Vincent's Hospital Clinical School, 1991) and the GlaxoSmithKline Advanced Trainee Research Award (Alfred Hospital, 2001). During the course of this research, Allen has been a recipient of a Postgraduate Medical Research Training Scholarship from the Australian National Health and Medical Research Council, the Murray Will Fellowship for Rural Physicians (Royal Australasian College of Physicians) and an Overseas Field Trip Award (Flinders University).

Publications

(i). Peer reviewed journal articles

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2. **Cheng A**, Nack Z, Graves S, McDonald M. Knowing where to look: "Meningococcal" septicaemia following a dog bite. Emergency Medicine, 1999 Sept; 11(3):185-187
3. **Cheng AC**, Winkel KD, Hawdon G, McDonald, M. "Irukandji-like" syndrome in Victoria. Australian and New Zealand Journal of Medicine, 1999 Dec; 29(6): 835
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8. Fowler VG, Olsen MK, Corey GR, Woods CW, Cabell CH, Reller LB, **Cheng AC**, Dudley T, Oddone EZ. Clinical identifiers of complicated *Staphylococcus aureus* bacteremia. Archives of Internal Medicine, 2003 Sept 22; 163: 2066-72
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11. **Cheng** AC, Mayo MJ, Gal D, Currie BJ. Chlorination and pH of drinking water do not correlate with rates of melioidosis in the Northern Territory, Australia. Transactions of the Royal Society of Tropical Medicine and Hygiene 2003; 97(5) 511-512
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16. Woods CW, **Cheng AC,** Fowler VG, Moorefield M, Frederick J, Sakoulas G, Meka VG, Tenover FC, Zwadyk FC, Wilson KH. Endocarditis caused by *Staphylococcus aureus* with reduced susceptibility to vancomycin. Clinical Infectious Disease 2004 Apr 15; 38: 1188 - 1191
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43. Maharjan B, Chantratita N, Vesaratchavest M, **Cheng AC**, Wuthiekanun V, Chierakul W, Chaowagul W, White NJ, Day NPJ, Peacock SJ. Recurrent melioidosis is often due to re-infection rather than relapse in patients in northeast Thailand. Journal of Clinical Microbiology (in press)

(ii). Other published work

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2. **Cheng AC**, Other people's practices; a world close to home. Medical Journal of Australia 1999; 170(7): 323-4.

3. **Cheng AC**, Ratcliff A, Adhikari P. Snake antivenom in Papua New Guinea. Fellowship Affairs, 2000 Jan; 19(1): 26

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10. **Cheng AC**. Emerging microbiologist. Microbiology Australia, 2004 July; 25(3): 43-44.
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(iii). Abstracts

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2. Winkel K, Hawdon G, **Cheng A**, Ashby K, Ozanne-Smith J, McDonald M. Jellyfish stings in Victoria, including an 'Irukandji-like' syndrome. Poster presentation at Annual Scientific Meeting of Australasian Society for Infectious Diseases and Australasian College of Tropical Medicine, Cairns, April, 1999.
3. **Cheng AC**, Mijch AM, Hoy JF, Wesselingh S, Fairley CK. The first 1000 days: An analysis of HIV-related discharges in the era of combination antiretrovirals. Poster presentation at Australian Society for HIV Medicine Annual Meeting, Perth, December 1999 [Abstract P-60]
4. **Cheng AC**, Mijch AM, Hoy JF. Myopathy, avascular necrosis and lipoatrophy in an HIV-infected male. Oral presentation at Australian Society for HIV Medicine Annual Meeting, Perth, December 1999 [Abstract OR-44]
5. Stevenson IH, **Cheng AC**, Hamer AWF. The significance of isolated troponin level elevations in acute rapid atrial fibrillation. Poster presentation at the Cardiac Society of Australia and New Zealand Annual Meeting, Melbourne, August, 2000.
6. Woods CW, **Cheng A**, Wilson K, Fowler V, Moorefield M, Tenover FC, Zwadyk P. Successful treatment of endocarditis caused by *Staphyloccoccus aureus* with reduced susceptibility to vancomycin with linezolid. Poster presentation at 1st International Symposium of Resistant Gram Positive Infections, San Antonio, December 3-5, 2000.

7. **Cheng AC**, Harrington G, Russo P, Liolios L, Spelman D. The effectiveness of isolation policies for the prevention of nosocomial VRE transmission; a quantitative study. Poster presentation at Society for Healthcare Epidemiology of America Annual Meeting, Toronto, April 1-3, 2001
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10. **Cheng AC**, Murdoch DR, Harrell LJ, Reller L*B.* Recurrent enterococcal bacteraemia; clinical and microbiological profile. Presentation at Australasian Society for Infectious Diseases Annual Meeting, Adelaide, April 13-17, 2002.
11. Stout JE, Fortenberry E, **Cheng ACS**, Engemann JJ, Hamilton CD. Safety and tolerability of the two-month regimen of rifampin and pyrazinamide for treatment of latent tuberculosis in Wake County, North Carolina. Presentation at the American Thoracic Society Annual Meeting, Atlanta GA, May, 2002.
12. **Cheng AC**, Jacups SJ, Currie BJ. A prognostic scoring system for melioidosis. Presentation at Australasian College for Tropical Medicine 11th Annual Scientific Meeting, Cairns, July 8-12, 2002
13. **Cheng A**, Stephens D, Anstey N, Fisher D, Currie *B.* G-CSF in the treatment of severe melioidosis; recent developments. Presentation at Australasian College for Tropical Medicine 11th Annual Scientific Meeting, Cairns, July 8-12, 2002
14. **Cheng AC**, Morahan G, Currie BJ, Browning GF. Responses of G-CSF knockout mice to *Burkholderia pseudomallei*. Presentation at Australian Society for Microbiology Annual Meeting, Melbourne, September 29-October 3, 2002. Microbiology Australia 2002 Sept 23(4) PP7.6,
15. **Cheng AC**, Currie BJ. A systematic review of G-CSF in the treatment of pneumonia. Presentation at the Royal Australasian College of Physicians (NT Committee) Annual Scientific Meeting, Darwin, November 16-17, 2002.
16. **Cheng AC**, Mayo MJ, Gal D, Foster N, Inglis TJJ, Currie BJ. Melioidosis and community water supply acidity. Presentation at Australasian Society for Infectious Diseases Annual Meeting, Canberra, March 22-26, 2003. Internal Medicine Journal 2003; 33(9-10): A69
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30. Anunnatsiri S, Chetchotisakd P, Mootsikapun P, White N and the **Melioidosis Study Group**. A comparison of ceftazidime versus combination ceftazidime-trimethoprim-sulphamethoxazole for the treatment of severe melioidosis. Oral presentation at 4th World Melioidosis Congress, Singapore, September 16-18, 2004

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