
UNIVERSITY OF NORTH TEXAS
HEALTH SCIENCE CENTER AT FORT WORTH
1970

CLINICAL INTERNSHIP IN THE SURGERY DEPARTMENT AT THE UNIVERSITY OF NORTH TEXAS HEALTH SCIENCE CENTER: ASSESSMENT OF HUMAN ANTIBODY RESPONSE TO UROKINASE PART A: A SPECIMEN ACQUISITION TRIAL FOR THE ASSESSMENT OF HUMAN ANTIBODY RESPONSE TO UROKINASE IN SUBJECTS TREATED FOR ACUTE LOWER-EXTREMITY ISCHEMIA

THESIS

Presented to the Graduate Council of the

Graduate School of Biomedical Sciences

University of North Texas

Health Science Center at Fort Worth

In Partial Fulfillment of the Requirements

For the Degree of

MASTER OF SCIENCE

By

Telicia A. Hughes, B.S.

Fort Worth, Texas

December 2003

CLINICAL INTERNSHIP IN THE SURGERY DEPARTMENT AT THE UNIVERSITY OF NORTH TEXAS HEALTH SCIENCE CENTER: ASSESSMENT OF HUMAN ANTIBODY RESPONSE TO UROKINASE

PART A: A SPECIMEN ACQUISITION TRIAL FOR THE ASSESSMENT OF HUMAN ANTIBODY RESPONSE TO UROKINASE IN SUBJECTS TREATED FOR ACUTE LOWER-EXTREMITY ISCHEMIA

Telicia A. Hughes, B.S.

APPROVED:

Rustin E. Reeves
Major Professor

Committee Member

Committee Member

James H. Alexander, PhD
University Member

Victoria Rudick
Graduate Advisor

Dean, Graduate School of Biomedical Sciences

ACKNOWLEDGEMENTS

First and foremost, I would like to thank God for granting me the strength, guidance and endurance to complete this master's program.

I would like to thank the entire Department of Surgery at the University of North Texas Health Science Center's Patient Care Center especially Della Weis and Chris Hayes for their help, support and guidance.

I would like to thank Dr. Rudick for all of her time, help and support.

I would also like to thank my family and friends for their support. I would like to especially thank my mother, father, and sisters for their help and encouragement and prayers.

Finally, most of all I would like to thank my son, Caleb Joseph Hughes for being my inspiration to endure the fight and overcome.

TABLE OF CONTENTS

TABLES

LITERATURE REVIEW

Urokinase (UK) is a proteolytic enzyme belonging to a class of agents known as plasminogen activators. These agents occur as both naturally produced enzymes (human and bacterial sourced) and as recombinant glycoproteins, capable of directly or indirectly converting the circulating human plasma pro-enzyme plasminogen into the active enzyme plasmin. Plasmin, in turn, is capable of dissolving fibrin, which is the predominant protein matrix in blood clots (thrombi). Abbokinase® is a human-sourced plasminogen activator manufactured by Abbott Laboratories and obtained from human neonatal kidney cells grown in tissue culture. The principal active ingredient in Abbokinase® is the low-molecular-weight form of urokinase, a direct plasminogen activator capable of converting plasminogen to plasmin.

Abbokinase® has been used for over 25 years for the treatment of several thromboembolic conditions. Abbokinase® was approved by the FDA in 1978 for intravenous use in the treatment of acute massive pulmonary embolism. This approval was primarily based on the results of two clinical trials sponsored by the National Heart and Lung Institute (NHLI). These two clinical trials, the Urokinase Pulmonary Embolism Trial (UPET)[1] and the Urokinase Streptokinase Pulmonary Embolism Trial (USPET)[2] showed a significant clinical benefit for intravenous urokinase treatment based on a decrease in pulmonary hemodynamic parameters. Although a human urinary based urokinase was used in these trials (high-molecular-weight urokinase isolated from human urine), a subsequent bridging study in pulmonary embolism, comparing urinary based

urokinase to human tissue-culture-sourced urokinase (Abbokinase®), showed the two products to be similar in efficacy and safety.[3] Abbokinase® has also been marketed for intracoronary use in the treatment of acute myocardial infarction and for treatment of occluded central venous catheters. Currently, Abbokinase® is approved and marketed for only the lysis of acute massive pulmonary emboli and for the lysis of pulmonary emboli accompanied by unstable hemodynamics.

Abbott Laboratories plans to assess the potential immunogenicity of Abbokinase® in humans in a population of subjects treated for acute lower-extremity ischemia. This patient population was chosen due to the relatively large range of doses utilized, and the extensive clinical experience with Abbokinase® in treating this disease.

Fibrinolysis

The existence of substances capable of fibrinolytic activity has been known for many years. In a review of the mechanisms of fibrin degradation (fibrinolysis), MacFarlane and Biggs[4] stated that over a hundred years earlier, Denis and Zimmerman had observed the dissolution of fibrin in human blood after standing for 12 to 24 hours. In 1933, Tillett and Garner[5] demonstrated that some strains of beta-hemolytic streptococci produced a substance that rapidly dissolved the fibrin in human plasma clots. This material was later found to be incapable of dissolving purified fibrin without the presence of a "lytic factor" associated with the euglobulin component of human serum[6] and was subsequently shown to be an activator for the proenzyme plasminogen, normally found in blood.[7,8]

Tillett and Sherry[9] ushered in the therapeutic use of fibrinolytic agents in 1949 when they injected concentrated, partially purified broth cultures of hemolytic streptococci into the pleural cavities of subjects suffering from various diseases that produced pleural exudates.

In 1947, MacFarlane and Pilling[10] described the fibrinolytic activity of normal urine, and in 1952, Astrup and Sterndorff[11] demonstrated that this activity was due to a plasminogen activator present in the urine. Sobel *et al*[12] designated the new plasminogen activator "urokinase" (UK). This material was a naturally occurring enzyme of human origin and as a result, had a major therapeutic advantage over the streptococcal derived material, streptokinase (SK). It is postulated to not induce the production of antibodies. Further more, UK, in contrast to SK, could be used in patients who have high streptococcal antibody titers as a result of recent streptococcal infections. Unfortunately, UK occurs only in trace amounts in urine. As a result, it was necessary to process large volumes of urine to obtain sufficient quantities of UK for clinical trials. For example, Lesuk *et al*[13] required 2300 liters of urine to isolate 29 mg of pure UK for determination of molecular weight.

The key to supplying adequate quantities of UK was discovered by Bernik and Kwaan[14,15] and Barlow and Lazer[16] who demonstrated that the enzyme could be obtained from human neonatal kidney cells grown in tissue culture. This information, along with the development of large-scale cell culture equipment by Weiss and Schleicher[17,18] is what prompted Abbott Laboratories to initiate a program to produce large quantities of

UK. Abbokinase® (urokinase) is a thrombolytic agent obtained from human kidney cells grown in tissue culture.

Although a number of investigators conducted small clinical studies to evaluate the use of two thrombolytic agents (UK and SK) in various disease states, the first large, controlled trial was the Urokinase-Pulmonary Embolism Trial (UPET)[19] organized and supervised by the National Heart and Lung Institute (NHLI). This trial, which compared the use of UK and heparin in the treatment of pulmonary embolism, was followed by another NHLI trial, the Urokinase-Streptokinase Pulmonary Embolism Trial (USPET)[20] which compared UK to SK. As a result to these trials, both UK and SK were approved by the FDA for use in pulmonary embolism in 1978.

Acute Lower-Extremity Ischemia

Approximately 60,000,000 Americans have cardiovascular disease, including one million Americans that are newly diagnosed with symptomatic peripheral vascular occlusive disease annually.[21] Atherosclerotic disease in the periphery is exceedingly common-the prevalence of plaque in the femoral arteries of patients over the age of 70 may be as high as 74%.[22] Many lesions are asymptomatic, although the incidence of symptomatic occlusion is significant, occurring in approximately 10% of patients greater than 65 years of age, and in more than 20% of patients 80 years and older.[23,24] The traditional treatment of chronic peripheral vascular disease is based on mechanical manipulation of occlusive lesions, with techniques such as percutaneous balloon angioplasty, stenting, surgical endarterectomy, and peripheral bypass. The clinical results of elective therapy are generally good, and 5-year patency and limb salvage rates range

from 50-80% depending on the anatomic site.[25,26] The results are less sanguine, however, for patients presenting with acute arterial ischemia (AAI). Patients with AAI typically present with severe pain, a range of sensory and neurologic deficits, and impending limb loss. The treatment can be notoriously challenging, given the limited time available to salvage ischemic tissue, the unknown vascular anatomy at the time of presentation, and the presence of thrombus in remote locations in the arterial system that are difficult to access by mechanical means. Because of the complexity and acuity of the disease, AAI continues to be a significant worldwide health problem with modern major amputation and mortality rates of nearly 20%.[27-29]

Generally, AAI results from either arterial thromboembolism, *in-situ* arterial thrombosis or graft thrombosis in patients with prior vascular reconstruction. Emboli most commonly originate from the left atrium in the presence of atrial dysrhythmias, but may occasionally arise from the left ventricle, thoracic or abdominal aorta (from mural irregularities or aneurysms), femoral artery aneurysms, superficial femoral atherosclerosis, popliteal aneurysms, of deep venous thrombus (DVT) via paradoxical embolism.[30] Emboli most commonly lodge in the common femoral (60%), aortoiliac (30%), and popliteal (10%) arteries.[27,30-32] AAI from *in-situ* thrombosis is less common than peripheral thromboembolism but carries a high rate of surgical failure, major amputation and death.[28,33,34] Thrombosis most often occurs in the setting of antecedent atherosclerosis of aneurysm, as the underlying lesions form a nidus for clot formation and propagation. Like atherosclerosis, *in-situ* thrombosis typically occurs at the superficial femoral (50%), aortoiliac (30%), and popliteal (20%) levels. *In-situ* thrombosis can also

occur in prior vascular grafts, a fairly common occurrence as only about 40% of infrainguinal autologous bypass grafts will remain patent after ten years.[35,36] The lack of available conduit, associated comorbidity, and the reoperative nature of these procedures make successful limb salvage problematic.

The traditional approach to patients presenting with AAI includes anticoagulation, surgical exploration and catheter thromboembolectomy.[27,28,31,32,37-40] These procedures are attended by high rates of complication, and are only marginally successful at restoring flow in patients with distal tibial and pedal artery thrombosis. The critical problem in management is to extract enough thrombus to both restore blood flow and to identify underlying lesion to plan for definitive vascular reconstruction. Extraction of segmental thrombi from large arteries (iliac, femoral) is fairly straightforward, but the ability to mechanically extract thrombi from distal outflow arteries (tibial, pedal) and arteriolar networks is limited.

A change in the management of AAI began in 1985 when McNamara and Fischer reported on a high-dose, local, intra-arterial regimen of urokinase that quickly reestablished blood flow through a totally occluded arterial segment with a minimum of complications.[41] Potential advantages of thrombolytic therapy include more exact anatomic localization (via angiography), and more gradual and complete clot dissolution and reperfusion.

The potential benefits of thrombolytic therapy in selected patients with AAI have been addressed in several clinical trials. The Rochester Trial, published in 1994, randomized 114 patients with AAI of less than seven days duration to receive either

urokinase (UK; Abbokinase®; Abbott Laboratories) or surgical thrombectomy.[37] The cumulative limb salvage rate was similar between the two groups (82% at 12 months) but the urokinase group demonstrated significantly reduced mortality (16% vs. 42% at 12 months; p=0.01). The STILE trial (Surgery vs. Thrombolysis for Ischemia of the Lower Extremity), conducted in 393 patients with both acute and chronic limb ischemia, was terminated early due to an observed significant benefit of initial surgery versus thrombolysis.[42] However, upon *post-hoc* stratification of the results by duration of ischemia (≤14 days vs. >14 days), a significant benefit was shown for the thrombolysis group in 6-month amputation free survival (6% vs. 18%) in the more acutely ischemic group (≤14 days). Additionally, a benefit was realized for those patients in whom successful thrombolysis lessened the severity of subsequent procedures. Amputation-free survival was maintained in 9% of the subjects whose procedure was lessened by at least one degree, as compared to a rate of 71% for surgical patients. Finally, the multicenter TOPAS trial (Thrombolysis or Peripheral Arterial Surgery) randomized 544 patients with AAI to receive either recombinant UK (r-UK) or surgery and revealed no differences in either the six-month amputation-free survival (72% vs. 75%) or mortality (16% vs. 12%), but found that 46% of patients treated with r-UK were discharged from the hospital alive without the need for open surgery.[43]

Molecular Weight

Urokinase consists of an A chain of 2,000 daltons linked by a single sulfhdryl bond to a B chain (active chain) of 30,400 daltons for a total molecular weight of approximately 32,000 daltons. (The two-dimensional AA sequence of Urokinase is provided in Appendix A, Figure 1.1.)

INTERNSHIP JOURNAL

June 2, 2003

9:00 A.M. Mrs. Della Weis and I reviewed the duties that I, as an intern, would be performing throughout the month of June. I received the protocol for a current study that is named Laparoscopic Cholecystectomy Using Ultrasonically Activated Scalpel Versus Monopolar Electrosurgery. I also received a protocol for the Urokinase study.

10:30 A.M. I met everyone on the 5th floor in the department of surgery. I also met everyone in the clinical trials department and the outpatient nurses.

11:30 A.M. I set up a tentative appointment to shadow a phlebotomist named Natalie at OMCT hospital for Monday, June 9, 2003. I will be creating my research proposal the week of June 9th due to Mrs. Della Weis's vacation. The research proposal is due June 22, 2003.

12:00 P.M. Lunch

1:00 P.M. I prepared my work space (Rm. 546). The Urokinase study will begin with an investigator's meeting in Chicago on June 27-29. (I will conduct my thesis based on this study). Friday, June 6, 2003, the closeout visit for the earwax study will be conducted.

2:30 P.M. Della constructed time in her schedule for me to be able to voice any concerns and or questions.

3:00-5:00 P.M. I became acquainted with the protocols of the Laparoscopic Cholecystectomy study and the Urokinase study.

June 3, 2003

9:00 A.M I became more acquainted with the Urokinase study's protocol.

12:00 P.M. Lunch

1:00-4:00 P.M. I did research on literature for my research proposal. I searched for literature containing information on lower extremity ischemia and thrombolytic agents such as Urokinase.

June 4, 2003

9:00 A.M. I performed research about Abbott Laboratories and the purpose for the Urokinase study.

12:00 P.M. Lunch

1:00-4:00 P.M. I did research on literature for my research proposal. I searched for more literature containing information on lower leg ischemia and thrombolytic agents such as Urokinase.

June 5, 2003

9:00 A.M. I checked in with Della to inform her of my arrival. She informed me of the work that I would be doing today. I received the protocol for the Tigecycline study by Wyeth Laboratories.

11:00 A.M. I went to the Clinical Trials Department to retrieve all of the elements of the Regulatory Binder for the Tigecycline study. I hole punched each sheet in order to fit the materials into the regulatory binder. Then, I separated the material into sections (protocol, CV, Financial Disclosure).

12:00 P.M. Lunch

1:00-5:00 P.M. I read some of the protocol for the Tigecycline study.

June 6, 2003

9:00 A.M. I checked in with Della to inform her of my arrival. She informed me of the work that I would be doing today. I voiced questions that I had accumulated from yesterday's assignment.

9:30 A.M. I continued organizing the Regulatory Binder for the Tigecycline study.

11:00 A.M. I read more of the Tigecycline study's protocol.

12:00 P.M. Lunch

1:00-5:00 P.M. I assisted in the closeout visit for the earwax study with Alcon of which Dr. Phillips is the Principal Investigator. The monitors went through all documents for the earwax study and flagged pages with questions for the Clinical Research Coordinator (Della). After we went through all of the documents, the monitor collected all packages and boxes of the drugs and leftovers.

June 9-June 16, 2003

I will be creating my research proposal this week due to Mrs. Della Weis's vacation.

June 17, 2003

9:00 A.M. I checked in with Della to inform her of my arrival. She informed me of the work that I would be doing today. I voiced questions that I had accumulated from Alcon's close out study and the protocols that I am reading.

11:00 A.M I assembled and organized the Safety Reports for the Linezolid vs. Vancomycin/Oxacillin/Dicloxacillin in the Treatment of Catheter-Related Gram Positive Bloodstream Infections study of which Dr. German Berbel is the Principal Investigator. The Safety Reports consists of the offsite SAE's (Serious Adverse Events). I had to summarize the events of the SAE and type the application for the SAE using a UNTHSC IRB (Institutional Review Board) application.

12:00 P.M. Lunch

1:00-5:00 P.M. I continued completing the Safety Reports.

June 18, 2003

8:30 A.M. I finished off the SAE applications for the Safety Reports. Then, I tried to find Dr. Berbel to sign the applications; however, he was not in the clinic at that time.

9:00 A.M. I checked in with Della to inform her of my arrival. She informed me of the work that I would be doing today. I voiced questions that I had accumulated from the Safety Reports.

10:00 A.M. Dr. Berbel signed the applications, and I mailed them to the IRB after making copies. I finished assembling the Tigecycline Regulatory Binder, and now I am assembling the Pharmacia Regulatory Binder for the Coronary Artery Bypass Graft study.

12:00 P.M. Lunch

1:00-5:00 P.M. I am continuing putting together the Pharmacia Regulatory Binder for the Coronary Artery Bypass Graft study. Then, I will work on my Research Proposal.

June 19, 2003

9:00 A.M. I checked in with Della to inform her of my arrival. She informed me of the work that I would be doing today.

10:00 A.M. I reread the Urokinase protocol so that I could be really familiar with the material.

12:00 P.M. Lunch

1:00 P.M. I went with Della to the Heart Place to set up an appointment with Dr. Wallace (Sub-Investigator) to meet with the monitor from Pharmacia/Pfizer (Sheila) on Monday June 23, 2003 for the initiation visit for the Coronary Artery Bypass Graft study.

2:30-5:00 P.M. I am reading the Medifacts International ECG Services Training Notebook.

June 23, 2003

9:00 A.M. I checked in with Della to inform her of my arrival. She informed me of the work that I would be doing today. I informed her of the procedures given in the Medifacts International ECG Services Training Notebook.

12:00 P.M. Lunch

1:00 P.M. The Pharmacia/Pfizer monitor (Sheila) is here for the initiation visit for the Coronary Artery Bypass Graft study. First, we will meet with Dr. Wallace, the Sub-Investigator of the study, at the Heart Place and discuss the study.

3:00-5:00 P.M. Sheila continued with the Coordinator section of the initiation visit and she performed an inventory of exactly what our site had received from Pharmacia/Pfizer.

June 24, 2003

8:30 A.M. I met Della in her office to get ready for the UNTHSC clinical trials meeting.

9:00 A.M The meeting began.

11:00 A.M I corrected Alcon queries.

12:00 P.M. Lunch

1:00-5:00 P.M. Della and I called Renee Ferguson at Medifacts International to schedule an ECG training, and I tried to do my online IRB training for the Coronary Artery Bypass Graft study, but the site is down. I went to clinical trials to pick up Dr. Yurvati's (the Principal Investigator of the Coronary Artery Bypass Graft study) 1572 and request the Pfizer's Financial Disclosure form for the Coronary Artery Bypass Graft study. Then, I completed and passed the tutorial for the IRB and received my certificate. Della and I discussed the schedule for tomorrow's site initiation visit with Wyeth for the Tigecycline study.

June 25, 2003

8:30 A.M. I am rereading some of the Tigecycline protocol in order to be completely prepared for the initiation visit today.

9:00 A.M. I checked in with Della to inform her of my arrival. She informed me of the work that I would be doing today. I voiced some questions that I had on the Tigecycline protocol.

10:00A.M.-5:00P.M. Brad Clinkscales, the monitor from Covance/Wyeth conducted a training/meeting with Dr. Berbel, Brenna LVN (Home Health Nurse), Della and I. Then he conducted another meeting with Della and me. Finally, we toured the pharmacy and microbiology department.

June 27-June 29, 2003

Dr. Yurvati, Della and I attended the investigator's meeting for the Urokinase study in Chicago.

June 30, 2003

8:30 A.M. I read some of the protocol for the Linezolid study in order to prepare myself for the initiation visit today which will be conducted by Sheila from Pfizer.

9:00 A.M. I checked in with Della to inform her of my arrival. She informed me of the work that I would be doing today. I voiced some questions that I had on the initiation visit yesterday.

11:00 A.M. We met with Sheila from Pfizer for the initiation visit of the Linezolid study. We engaged in a brief discussion about the study.

12:00 P.M. Lunch

1:00-5:00 P.M. Sheila then conducted the initiation visit.

July 1-July 4, 2003

Della is on vacation; therefore, I will work on my research proposal.

July 7, 2003

9:00 A.M. I checked in with Della to let her know that I am here. She informed me of the work that I would be doing today.

10:00 A.M. I am reading the Coronary Artery Bypass Graft study's protocol.

12:00 P.M. Lunch

1:00 P.M. I am still reading the Coronary Artery Bypass Graft study's protocol.

4:00-5:00 P.M. I am working on my research proposal, and the signoff sheets for the research proposal.

July 8, 2003

11:00 A.M. I checked in with Della to inform her of my arrival. She informed me of the work that I would be doing today. Della and I got the pharmacy to sign the delegation sheet for the Coronary Artery Bypass Graft study, and brought Dr. Wallace his Financial Disclosure sheet.

12:30 P.M. Randy Avers called and confirmed to do site initiation for Urokinase on Thursday, July 10, 2003 at 1:00 P.M. The ECG training for CORONARY ARTERY BYPASS GRAFT is on Tuesday, July 9, 2003 at 12:00 P.M.

1:00-5:00 P.M. I will continue to work on my research proposal and read more of the Coronary Artery Bypass Graft study's protocol.

July 9, 2003

9:00 A.M. I checked in with Della to inform her of my arrival. She informed me of the work that I would be doing today. I voiced some questions that I had on the Coronary Artery Bypass Graft study's protocol.

10:45 A.M. I read the Coronary Artery Bypass Graft study's newsletter and more of the protocol. Della and I are supposed to have the ECG training today at 12:00 noon.

12:00 P.M. Della and I had the ECG training, it was very long and extensive.

3:00-5:00 P.M. I finished my draft to my research proposal. Yet, I have many questions for Dr. Reeves and Dr. Rudick. We are anticipating the site initiation visit with Randy Avers tomorrow.

July 10, 2003

9:00 A.M. I checked in with Della to inform her of my arrival. She informed me of the work that I would be doing today. I voiced some questions that I had about my research proposal.

11:00 A.M. I helped Della put together an itemized list of all the studies that the department of surgery is conducting and how much each study is paying. We are waiting on Abbott's monitor after giving a letter to the Microbiology Department and Pharmacy, and Dr. Byers about the Linezolid study.

1:00 P.M. The Abbott monitor and an associate visited for the site initiation visit of the Urokinase study.

4:00-5:00 P.M. I conversed with Della about the nature of a coordinating position.

July 11, 2003

9:00 A.M. I checked in with Della to inform her of my arrival. She informed me of the work that I would be doing today.

10:00 A.M. We are now organizing the Coronary Artery Bypass Graft and the Linezolid study's CRFs (Case Report Forms). Then, Della and I registered into the ClinPhone IVRS (Interactive Voice Response System) for the Coronary Artery Bypass Graft study.

12:00 P.M. Lunch

1:00 P.M. Della and I tried to register into the ClinPhone IVRS (Interactive Voice Response System) for the Linezolid study; however, the system is not responding to our site's investigator's number. ****Remember to tell Sheila on Monday that the Investigator's number for this site is incorrect therefore we were not able to complete the IVRS registration for the Linezolid study. IVRS is according to FDA 21 CFR Part11 Certification (Provision of IVRS Access codes). FDA 21 CFR Part 11 is the local regulation governing the use of computerized systems for Clinical Research protocols.

3:00-5:00 P.M. I read more of the Linezolid and Coronary Artery Bypass Graft study protocols. On Monday we have an in-service meeting with Sheila Easely (the monitor for Pfizer) for the Linezolid study. In this in-service meeting copies of the protocol synopsis, and Inclusion/Exclusion criteria were distributed. The protocol was discussed, and we came up with a recruitment strategy. For the Linezolid study, we would identify all patients with a central

catheter and attach stickers to alert the house staff and caretakers that if they suspect an infection call the Principal Investigator {Dr. Berbel}, Della, or Telicia. Also, we alert the staff to please do not pull the catheter before the culture is taken. The informed consent was discussed. We made sure that the informed consent is available in the Pharmacy, Microbiology, and other places accessible to all staff involved in the study. We must issue a copy of the schedule of events for protocol procedure {collect 2 cultures, peripheral and catheter}. By Monday, reread the Linezolid and Urokinase protocols, really be familiar with both because Urokinase will probably enroll on Tuesday.

July 14, 2003

9:00 A.M. I checked in with Della to inform her of my arrival. She informed me of the work that I would be doing today.

10:00 A.M. Della and I began conducting the in-service to the residents and the microbiology staff. The Pfizer monitor will come later (Sheila arrived at 10:30). We issued the protocol synopsis and I&E (Inclusion/Exclusion) cards to the residents. Then, we discussed the protocol with Larry the Microbiologist and the residents. Sheila and I will meet in August and ******remind her to check the Linezolid drugs in the Pharmacy because they expire in August.

2:30 P.M. Della and I had a discussion with the Pfizer monitor about the Linezolid study and the IVRS problem.

4:00-5:00 P.M. I read more of the Linezolid protocol.

July 15, 2003

9:00 A.M. I checked in with Della to inform her of my arrival. She informed me of the work that I would be doing today. Now we are ready to enroll in all studies except the Linezolid study because Larry in Microbiology has not received the study plates.

10:00 A.M. I called Dr. Wallace's office to check if there are any Coronary Artery Bypass Graft patients scheduled.

12:00 P.M. Lunch

1:00-5:00 P.M. I worked on research proposal and the form for designation of committee, and read more of the Coronary Artery Bypass Graft study's protocol. I also performed a personal search about Coronary Artery Bypass Grafts.

July 16, 2003

9:00 A.M. I checked in with Della to inform her of my arrival. She informed me of the work that I would be doing today.

10:00 A.M. I typed some of my research proposal.

12:00 P.M. Lunch

1:00-5:00 P.M. I typed most of my research proposal and read Coronary Artery Bypass Graft, urokinase protocols. Della and I performed mock trials of conducting informed consent.

July 17, 2003

9:00 A.M. I checked in with Della to inform her of my arrival. She informed me of the work that I would be doing today. I am waiting on Dr. Reeves to e-mail me back with the revisions on my research proposal and CV (Curriculum Vitae).

11:00 A.M. I am working on my research proposal and CV.

12:00 P.M. Lunch

1:00-4:00 P.M. I am working on my research proposal and CV. I will also read more of the Tigecycline study's protocol.

July 21, 2003

9:00 A.M. I checked in with Della to inform her of my arrival. She informed me of the work that I would be doing today.

10:00 A.M. I went to register for the 5600 core course. I also checked for a response from Dr. Reeves on my research proposal.

12:00 P.M. Lunch

1:00 P.M. We enrolled a patient for the Coronary Artery Bypass Graft study today. We saw Sheila Ford, R.N. in the hallway at the hospital and she informed us of a potential Coronary Artery Bypass Graft patient. Then, we went to Reggie in Outpatient Registry to see if

	the patient had arrived. We talked to the patient and the patient's family and the patient agreed and met all of the inclusion and none of the exclusion criteria. Then, the patient signed the informed consent; then we collected the ECG; then we collected the patient's blood work.
4:00-5:00 P.M.	We will now transmit the ECG to Core Labs. Dr. Wallace may have two more patients tomorrow. We processed and sent off the blood work to Covance.

July 22, 2003

9:00 A.M.	I checked in with Della to inform her of my arrival. She informed me of the work that I would be doing today.
10:00 A.M.	I filled out the SAE applications for UNTHSC's IRB for more offsite safety reports for the Tigecycline study.
12:00 P.M.	Lunch
1:00 P.M.	I worked on my research proposal
2:00 P.M.	Della and I met Ron Como (Director of ICU) to inform him and make him aware of the study being performed and on what patient. We also brought Ron a copy of the protocol synopsis for the CABG study.
3:30 P.M.	I looked for Dr. Berbel to sign the SAE applications to the IRB. I also made labels to go on the patients' chart to alarm the staff of a CABG study patient.

4:00-5:00 P.M. We made copies of the Coronary Artery Bypass Graft patient's urinalysis and CBC. We will meet in the OR (Operating Room) at 9:00 tomorrow, wear tennis.

July 23, 2003

9:00 A.M. I checked in with Della to inform her of my arrival. She informed me of the work that I would be doing today.

9:30 A.M. Della and I are headed to the OR for the first Coronary Artery Bypass Graft patient's bypass. Patient 1 should be extubated around 5:00 P.M.; therefore, we will randomize patient 1 at that time.

1:00-6:00 P.M. We screened the second patient and the patient's operation is tomorrow on the July 24, 2003. There is another patient scheduled to be screened on the July 30, 2003.

July 24, 2003

9:00 A.M. I checked in with Della to inform her of my arrival. She informed me of the work that I would be doing today.

10:00 A.M. I sent the Covance labs and added dry ice. I helped patient 1 with the patient diary this morning. We are performing the first patient's Day 1 for the study and the second patient's Surgery Day/Baseline (since Dr. Wallace does his extubations on the same day).

12:00 P.M. Lunch

1:00 P.M. I went to check on the first patient, and I visited with the patient's family and spoke to Sheila Ford, R.N. about the surgery and the study.

3:30-5:00 P.M. I read more on the Coronary Artery Bypass Graft study's protocol.

July 25, 2003

8:00 A.M. I went to perform both the first and second patient's ECG before drug administration. The second patient was too sick to perform the ECG.

11:00 A.M. We took the second patient's ECG and randomized that patient.

12:30 P.M. Lunch

1:30-4:00 P.M. We helped each patient fill out their diary. We made copies of the first patient's medical records. We visited with both patient's families.

July 26, 2003

9:00 A.M. I checked in with Della to inform her of my arrival. She informed me of the work that I would be doing today.

10:00 A.M. I went to the hospital to check on the patients.

11:00 A.M. I performed the second patient's ECG.

12:00 P.M. Lunch

1:00 P.M. I helped both patients complete their personal diary.

3:30-5:00 P.M. I read more on the Urokinase and Tigecycline study.

July 27, 2003

7:30 A.M.	I arrived and met with the second patient's family and performed the second patient's ECG.
10:00 A.M.	I then transmitted all of the ECGs to Medifact's Core Lab.
12:00-1:00 P.M.	I then helped both patients complete their personal diary.

July 28, 2003

9:00 A.M.	I checked in with Chris (new hire/PA) to inform him of my arrival. Della will be on vacation.
10:00 A.M.	I performed the first patient's ECG and helped both patients complete their personal diary. I had the labs drawn for both patients.
12:00 P.M.	Lunch
1:00 P.M.	I helped process the blood work of both patients and fill out the CRFs.
1:30 P.M.	I sent off the labs to Covance and went to the Outpatient Registry at the Osteopathic Medical Center to enroll the third patient.
4:30-5:00 P.M.	We copied all patients' medical charts that were enrolled in the CABG study.

July 29, 2003

9:00 A.M.	The first patient is discharged. I went to check on the third patient and to speak to the patient's spouse who seems very skeptical.

11:00 A.M. The third patient will be noted as a screen failure because the patient was extubated an hour late.

12:00 P.M. Lunch

1:00 P.M. We called ClinPhone and characterized the patient as a screen failure. There are two more prospective patients for the Coronary Artery Bypass Graft study. We notified the patient's spouse who was very understanding.

3:00-5:00 P.M. I worked on the first patient's CRF and the second patient's narrative.

July 30, 2003

9:00 A.M. I went to check on our Coronary Artery Bypass Graft patients. The second patient is still not discharged and will be held another day because of the patient's mental status.

11:00 A.M. I worked on the first and second patient's CRFs of the CABG study. We went to visit the third patient although that patient was characterized as a screen fail patient.

12:00 P.M. Lunch

1:00-5:00 P.M. We copied more of the second patient's charts. I worked on more of the first patient's CRF and transmitted all of the ECGs to Medifact's Core Lab.

July 31, 2003

9:00 A.M.	I went to visit the second patient who seems to be scared to go home. I was notified that the patient will be discharged on Saturday.
11:00 A.M	I performed an ECG on the patient.
12:00 P.M.	Lunch
1:00-5:00 P.M.	I worked on both patient's CRFs and transmitted the ECG to Medifact's Core Lab.

August 1, 2003

8:00 A.M.	We consented the first Urokinase patient.
8:30 A.M.	We performed all necessary pre-treatment assessments including collection of medical history and blood work. The blood work was collected in four red top tubes.
9:00 A.M.	We got the drug from the pharmacy and watched Dr. Peska inject the thrombolytic agent. I brought the red top tubes of blood to microbiology to be centrifuged.
10:00 A.M.	Then, we processed the centrifuged blood and aliquoted the blood in to 5-1ml vials. Then, we put the specimens in the –70 freezer. Then, we faxed and called Randy about the first Urokinase patient.
1:00 P.M.	Lunch
2:00-5:00 P.M.	I read the Urokinase protocol and sorted through the two Coronary Artery Bypass Graft patient's source documents.

August 5, 2003

9:00 A.M.	I sorted through the two Coronary Artery Bypass Graft patient's source documents.
12:00 P.M.	Lunch
1:00 P.M.	We saw the first and second patient of the Coronary Artery Bypass Graft study for their final visit. They are both doing really well. We collected both patient's ECGs and blood work.
4:00-5:00 P.M.	We processed the blood work and shipped that blood work off to Covance for analysis. We deleted ECGs per Medifacts.

August 6, 2003

9:00 A.M.	We went to visit the third patient.
10:00 A.M.	We placed labels on the first Urokinase patient's specimens.
10:30 A.M.	We collected information for the Charisma study.
12:00 P.M.	Lunch
1:00-5:00 P.M.	We put together some of the source documents and we contacted Randy Avers to see when he was going to visit and monitor. We completed some of the CRFs for the Urokinase patient.

August 7, 2003

9:00 A.M.	I worked on the narrative for the second Coronary Artery Bypass Graft study's patient and the first Urokinase patient's CRF.
12:00 P.M.	Lunch

1:00-5:00 P.M. I worked on the Urokinase patient's source documents. I deleted all ECGs for the Coronary Artery Bypass Graft study per Medifacts. I completed my research proposal.

August 8, 2003

9:00 A.M. I turned in my research proposal.

10:00 A.M. I organized the screen failures with Medifacts over the phone.

12:00 P.M. Lunch

1:00 P.M. I met with a fifth Coronary Artery Bypass Graft study patient. The patient agrees to participate and meets all of the pre-operative inclusion and none of the pre-operative exclusion criteria. I performed an ECG and arranged for the blood work to be collected from the patient.

4:00-5:00 P.M. We processed the blood work and shipped that blood work off to Covance for analysis.

August 11, 2003

9:00 A.M. I prepared all of the necessary paperwork for the Coronary Artery Bypass Graft study's fifth patient because that patient will be randomized tomorrow.

11:00 A.M. I gathered all of the Urokinase information and assembled it into the Regulatory Binder.

12:00 P.M. Lunch

1:00-5:00 P.M. I continued assembling the Regulatory Binder. I then worked on the Urokinase study's first patient's source documents. I assembled binders for all patients' source documents and all screen failures. The Abbott monitor acknowledged that he will visit our site Wednesday, August 20, 2003 at 9:00 A.M.

August 12, 2003

7:30 A.M. We examined all of the Coronary Artery Bypass Graft study's fifth patient post-operative inclusion/exclusion criteria. The patient met all of the inclusion and none of the exclusion criteria.

8:00 A.M We went to the pharmacy and called ClinPhone to randomize the patient. Once, the patient was randomized, we notified the pharmacy and the study drug was sent to the floor.

9:00 A.M. I went to class.

11:00 A.M. I performed and ECG on the patient. I arranged for the patient's blood to be collected. I assessed the patient's vital signs. Another Coronary Artery Bypass Graft patient is pre-oping for surgery and I went to the outpatient to inform this patient as a candidate for the study.

1:00 P.M. The patient did not arrive.

1:20 P.M. We met the first Coronary Artery Bypass Graft study's patient at the Heart Place for a retest because the patient's initial labs for the

	final visit were elevated and a retest was requested per Principal Investigator. We collected the patient's blood.
3:30-4:30 P.M.	We processed the first and fifth Coronary Artery Bypass Graft patients blood and shipped the blood work to Covance.

August 13, 2003

9:00 A.M.	We organized all CRFs and binders into storage and locked cabinets.
12:00 P.M.	Lunch
1:00 P.M.	I started the Coronary Artery Bypass Graft study's fifth patient's CRF, and I finished up the Coronary Artery Bypass Graft study's second patient's narrative. I called Medifacts and request more electrodes and more ECG paper.
3:45-5:00 P.M.	We drew the fifth patient's blood work and collected that patient's ECG. We then processed the fifth patient's blood work and called Covance to retrieve that blood work.

August 14, 2003

9:00 A.M.	I went to class.
11:00 A.M.	We collected the fifth patient's ECG and blood work and processed that blood work. I began to write in the fifth patient CRFs.
12:00 P.M.	Lunch

1:00 P.M. I contacted Randy, Abbott's monitor, and gave him the information for Wednesday night (Abbott treated our site out to dinner because we were the first to successfully enroll in the Urokinase study).

2:00 P.M. I checked with Dr. Wallace to see if he had completed the fifth patient's NIH stroke scale, and issued to him the first patient's laboratory results. The patient's lab results may reveal an AE (Adverse Event).

4:00-5:00 P.M. I typed the second patient's narrative.

August 15, 2003

9:00 A.M. We collected blood work for the fifth patient's IV/PO switch day. Today, the fifth patient switched to oral medication. We collected more of the patient's history, copied records for (the Urokinase study's first patient and the Coronary Artery Bypass Graft study's fifth patient). We also collected some of the Coronary Artery Bypass Graft study's fifth patient's personal diary.

1:00 P.M. Lunch

2:00 P.M. We completed more of the CRFs for the Coronary Artery Bypass Graft and Urokinase studies. I e-mailed ClinPhone about the situation for screen failure for the Coronary Artery Bypass Graft study's fourth patient.

4:00-5:00 P.M.	We processed and shipped the blood work of the fifth patient to Covance. I organized source documents of the Coronary Artery Bypass Graft study's patients.

August 18, 2003

9:00 A.M.	I went to 5600 class.
11:00 A.M.	I transmitted all untransmitted ECGs
12:00 P.M.	Lunch
1:00 P.M.	I discussed the Coronary Artery Bypass Graft CRFs with Pfizer's monitor Sheila Easley.
3:00 P.M.	The Coronary Artery Bypass Graft study's fifth patient is being discharged. I collected the patient's ECG and some of the patient diary. I informed the patient on how to take the medications and what to bring for the final visit.
4:30-5:00 P.M.	I transmitted the fifth patient's ECG.

August 19, 2003

9:00 A.M.	I organized all of the Urokinase study's material (CRFs, Source Documents, Regulatory Binder, etc....)
1:00 P.M.	I went to my appointment with Dr. Reeves.
2:30 P.M.	I cleaned my office space for the Abbott monitor coming tomorrow.
3:00-5:00 P.M.	I self-monitored the Urokinase patient's CRF and Source Documents.

August 20, 2003

8:00 A.M. I checked in with Della, and we discussed the things that Abbott's monitor may need and request. I went through CRFs before the Abbott monitor arrived.

9:00 A.M. Monitor checked CRFs, source documents, regulatory binder, etc...

1:00 P.M. We discussed the changes that needed to be made and the documents that needed to be added.

1:30 P.M. We escorted the monitor to the pharmacy where he checked on the drug inventory. On August 22 there is another Coronary Artery Bypass Graft patient to enroll.

2:00-5:00 P.M. The Abbott monitor left and I continued correcting all things that needed to be corrected. I assembled a Pre-operative history sheet for the Coronary Artery Bypass Graft study so that it will be easier for us to collect the patient's history.

7:00-9:00 P.M. We arrived to dinner at Texas de Brazil. Dr. Peska notified us that the Urokinase study's first patient will need to have an Aortal Bi-femoral surgery because his legs hurt, and that may be an SAE for the Abbott study.

August 21, 2003

8:00 A.M. I reviewed the forms that needed to be completed for an SAE for the Urokinase study.

9:00 A.M. I went to class.

11:00 A.M. I discussed the SAE situation with Della.

12:00 P.M. Lunch

1:00-5:00 P.M. I tried to stay current with the first Urokinase patient's Source Documents/Medical Charts.

August 22, 2003

9:00 A.M. I called Abbott's Medical Monitor about the SAE and she stated that the Aortal Bi-femoral surgery would not be considered and SAE, it is and AE.

12:00 P.M. Lunch

1:00 P.M. We conducted an informed consent to the Coronary Artery Bypass Graft's sixth prospective patient. The patient accepted and is scheduled to have surgery on Mon., Aug. 25, 2003. I copied the Urokinase patient's chart for the surgery day.

3:00-5:00 P.M. We processed the Coronary Artery Bypass Graft's sixth patient's blood work and shipped the blood work to Covance. I transmitted the sixth patient's ECG. We conducted and informed consent for the Tigecycline study; however, the patient turned out to be a screen fail because the patient developed pseudomonas.

August 25, 2003

9:00 A.M. I went to check on the Coronary Artery Bypass Graft study's sixth patient after the surgery and I asked the CCU nurse to page me once the patient is extubated.

10:00 A.M. I called Abbott's Medical Monitor about the AE and she stated that the Aortal Bi-femoral surgery is not even and AE because Abbott is FDA exempt for those type of surgeries, so no paperwork is needed.

12:00 P.M. Lunch

1:00 P.M. I tried to initiate the ECG machine for the Tigecycline study. I contacted Renee from Medifacts to delete ECGs.

2:00-5:00 P.M. I learned that the Urokinase study's first patient endured an SAE over the weekend. I contacted Abbott's personnel to inform them of the SAE. The patient had endured compartment syndrome leading to a fasciotomy. I collected Medical Charts and reviewed the subsequent paperwork.

August 26, 2003

9:00 A.M. I went to class.

11:00 A.M. I filled out the SAE forms for the Urokinase study's first patient.

12:00 P.M. Lunch

1:00 P.M. We screened the Coronary Artery Bypass Graft study 's seventh patient. The patient met all criteria and agreed to participate. I collected the seventh patient's ECG and arranged for the seventh patient's blood work to be collected. We went to the Heart Place and met the Coronary Artery Bypass Graft study 's fifth patient for

	the final visit. We collected the fifth patient's blood work and ECG.
3:00-5:00 P.M.	We collected an ECG and Blood work for the Coronary Artery Bypass Graft study 's sixth patient. I copied more of the Urokinase patient's chart. We processed and shipped all of the Coronary Artery Bypass Graft study's patients' blood work.
9:00 P.M.	The Urokinase second patient was informed and enrolled.

August 27, 2003

9:00 A.M.	I filled out more of the SAE forms. I am only waiting for Dr. Peska's narrative, medications and supplements, and non-protocol sheet.
12:00 P.M.	Lunch
1:00 P.M.	I sent SAE (Serious Adverse Event) forms to Abbott and our IRB. I deleted ECGs for Medifacts, and ordered a new ECG machine. I collected the sixth patient's ECG, blood work and diary.
4:00 P.M.	I processed the sixth patient's blood work and shipped the blood work to Covance.

August 28, 2003

9:00 A.M.	I went to class.
11:00 A.M.	I called Renee at Medifacts about the broken ECG and we received a new machine. I transmitted all ECGs in the old machine.
12:00 P.M.	Lunch

1:00-5:00 P.M. I went to Charisma study's site initiation visit.

August 29, 2003

8:00 A.M. We randomized the seventh patient of the Coronary Artery Bypass Graft study. We collected the patient's ECG, blood work, and helped the patient with the diary. We helped the sixth patient of the Coronary Artery Bypass Graft study with the diary, and reminded the patient to bring a list of the concomitant medications being taken.

12:00 P.M. Lunch

1:00-5:00 P.M. I copied the Urokinase study's first and second patient's charts. I arranged for the Urokinase study's second patient's blood work to be collected (third day assessment). We processed Coronary Artery Bypass Graft study's sixth and seventh patient's blood work. I faxed Medifacts a letter stating the Coronary Artery Bypass Graft study's fourth patient's (screen failure) birthday.

August 30, 2003

7:30 A.M. I collected Coronary Artery Bypass Graft study's seventh patient's ECG and arranged for the patient's blood work to be collected. I helped fill out the patient diary with the patient. I informed Coronary Artery Bypass Graft study's sixth patient to bring the conmeds. I also reconfirmed the patient's discharge on 8/31/03.

I helped the sixth patient with the patient diary. I explained the importance of compliance with the patient diary.

10:00-11:00 A.M. I processed all blood work and shipped off the ambient specimens to Covance and put the CK/CKMB specimen in the –70 freezer.

August 31, 2003

8:00 A.M. I collected the Coronary Artery Bypass Graft study's seventh patient's ECG and blood work. I collected the Coronary Artery Bypass Graft study's sixth patient's ECG and assisted with the diary. I assisted with the diary for the seventh patient also.

10:00 A.M. I reinforced compliance with the patient diary to the sixth patient. I explained to the sixth patient about the final visit and what material to bring to the final visit. The patient agreed and understood.

10:30-11:20 A.M. I processed all blood work and put the ambient specimens in the refrigerator and the frozen specimens in the -70 freezer.

September 1, 2003

8:00 A.M. I collected the seventh patient's blood work. I tried to locate the seventh patient's oral dose (Today is IV/PO switch for this patient). I paged Della to inform her of the missing medication and she contacted the pharmacy and requested that the pharmacy dispense another pill until the other one is found.

10:30-11:00 A.M. I processed all blood work, and I put the ambient specimens in the refrigerator and the frozen specimens in the -70 freezer.

September 2, 2003

9:00 A.M. I went to class.

11:00 A.M. I worked on Abbott's CRFs for the second Urokinase patient.

12:00 P.M. Lunch

1:00 P.M. I met the Coronary Artery Bypass Graft study's first patient for the 30-day follow-up. We collected the patient's ECG, vitals, and blood work. We thanked the patient for participating in the study, and we saw the fifth patient in the lobby of the Heart Place. We informed the patient to mail the materials that the patient forgot to bring.

4:00 -5:00 P.M. We processed the first patient's blood work and transmitted the ECG.

September 3, 2003

9:00 A.M. I worked on the Urokinase study's second patient's CRF.

12:00 P.M. Lunch

1:00 P.M. The Coronary Artery Bypass Graft study's second patient arrived for the 30-day follow up. We collected the patient's ECG and bloodwork. The seventh patient was discharged. I informed the patient about compliance with the personal diary and what

	materials to bring to the final visit. The patient agreed and understood.
4:00-5:00 P.M.	We processed the second patient's blood work and shipped the specimens to Covance.

September 4, 2003

9:00 A.M.	I went to class.
11:00 A.M.	I worked on all Abbott CRFs.
12:00 P.M.	Lunch
1:00 P.M.	I worked on all Abbott CRFs.

September 5, 2003

9:00 A.M.	I worked on all Abbott CRFs and Source Documents.
12:00 P.M.	Sheila (Pfizer monitor) took us out to lunch for good work on the Coronary Artery Bypass Graft study.
1:00 P.M.	I scheduled the Urokinase study's second patient's appointment.
1:30 P.M.	I worked on all Abbott CRFs.

September 8, 2003

9:00 A.M.	I worked on all Abbott CRF and Source Documents. I copied charts for the third Urokinase patient.
12:00 P.M.	Lunch

1:00 P.M. I handled kinks with the first patient's Source Documents with the Abbott monitor over the phone. We also discussed the Abbott protocol for the Urokinase.

2:00 P.M. I worked on all Abbott CRFs.

September 9, 2003

9:00 A.M. I went to class.

11:00 A.M. I worked on all Abbott CRFs and Source Documents.

12:00 P.M. Lunch

1:00 P.M. The Coronary Artery Bypass Graft study's sixth and seventh patient arrived for their 30-day follow-up and we collected their blood work, vitals, and ECGs. I transmitted the Coronary Artery Bypass Graft study's sixth and seventh patient's ECGs. The Urokinase study's second patient's third blood draw is tomorrow.

3:30 P.M. We processed the sixth and seventh patient's blood work and shipped the specimens to Covance.

4:30-5:00 P.M. I am preparing necessary questions for the Urokinase study's second patient's follow-up tomorrow.

September 10, 2003

9:00 A.M. I worked on CRFs and Source Documents for all Urokinase study patients.

12:00 P.M. Lunch

1:00-5:00 P.M. I resolved questions on the Coronary Artery Bypass Graft study's seventh patient with Medifacts. I continued to work on CRFs and Source Documents for all Urokinase study patients.

September 11, 2003

9:00 A.M. I went to class.

11:00 A.M. I worked on CRFs and Source Documents for all Urokinase study patients.

12:00 P.M. Lunch

1:00-5:00 P.M. I continued to work on CRFs and Source Documents for all Urokinase study patients.

September 12, 2003

9:00 A.M. I worked on CRFs and Source Documents for all Urokinase study patients.

12:00 P.M. Lunch

1:00-5:00 P.M. I continued to work on CRFs and Source Documents for all Urokinase study patients.

September 13, 2003

9:00 A.M. I worked on CRFs and Source Documents for all Urokinase study patients.

12:00 P.M. Lunch

1:00-5:00 P.M. I continued to work on CRFs and Source Documents for all Urokinase study patients.

September 15, 2003

9:00 A.M.	I worked on CRFs and Source Documents for all Urokinase study patients.
12:00 P.M.	Lunch
1:00-5:00 P.M.	I continued to work on CRFs and Source Documents for all Urokinase study patients.

September 16, 2003

9:00 A.M.	I went to class.
11:00 A.M.	I worked on CRFs and Source Documents for all Urokinase study patients.
12:00 P.M.	Lunch
1:00-5:00 P.M.	I continued to work on CRFs and Source Documents for all Urokinase study patients.

September 17, 2003

9:00 A.M.	I worked on CRFs and Source Documents for all Urokinase study patients.
12:00 P.M.	Lunch
1:00-5:00 P.M.	I continued to work on CRFs and Source Documents for all Urokinase study patients.

September 18, 2003

9:00 A.M.	I went to class.

11:00 A.M. I met the Urokinase study's third patient for the third antibody assessment blood draw. I worked on CRFs and Source Documents for all Urokinase study patients.

12:00 P.M. Lunch

1:00-5:00 P.M. I continued to work on CRFs and Source Documents for all Urokinase study patients.

September 19, 2003

9:00 A.M. I worked on CRFs and Source Documents for all Urokinase study patients. The Abbott monitor will come to our site to monitor on Wednesday September 24, 2003.

12:00-1:00 P.M. The eighth Coronary Artery Bypass Graft patient was enrolled.

September 22, 2003

9:00 A.M. I worked on CRFs and Source Documents for all Urokinase study patients.

12:00 P.M. Lunch

1:00-5:00 P.M. I continued to work on CRFs and Source Documents for all Urokinase study patients.

September 23, 2003

9:00 A.M. I went to class.

11:00 A.M. I worked on CRFs and Source Documents for all Urokinase study patients.

12:00 P.M. Lunch

1:00-5:00 P.M. I continued to work on CRFs and Source Documents for all Urokinase study patients. The Urokinase study's second patient's last antibody assessment blood draw is scheduled for September 24, 2003.

September 24, 2003

9:00 A.M. The Urokinase study's second patient called to reschedule the appointment.

9:30 A.M. The Tigecycline study's monitor arrived and monitored the CRF and Source for the screen fail patient. The Tigecycline study's monitor was very pleased with our site and offered this site another study in collaboration with the Tigecycline study.

12:00 P.M. Lunch

12:30 P.M. I am preparing for the Abbott monitor to arrive at 1:00 P.M..

1:00-5:30 P.M. Randy (the Abbott monitor) arrives, and he and I went through all CRFs and Source Documents while acknowledging and correcting mistakes. We will continue going through source documents tomorrow.

September 25, 2003

8:00 A.M. I tried to make more corrections to the CRFs that were requested by Randy.

9:00 A.M. Randy arrives, and he and I went through all CRFs and Source Documents.

10:00 A.M. The Urokinase study's second patient arrived for the last antibody assessment blood draw. I asked the patient necessary questions regarding the patient's medical history. We collected the patients blood work, and Dr. Delange saw the patient.

12:00 P.M. Lunch

1:00 P.M. Della and Chris went through particular CRFs and Source Documents with Randy. The monitor was not able to finish monitoring everything due to the volume of material. He will have to come back to monitor. We set up a tentative date for the monitor to come back to our site. The date is as follows: October, 14 and 15, 2003.

3:30 P.M. Della walks the monitor over to the pharmacy to view drug inventory. Chris and I separate the Source Documents into sections (Physician's Notes, Operation Reports, Nurse's Notes, Nurse's Flowcharts, etc.....)

5:00-5:30 P.M. The Coordinating Team discussed the monitor's visit.

September 26, 2003

9:00 A.M. We collected the eighth Coronary Artery Bypass Graft study's patient's ECG and blood work (IV/PO switch).

12:00 P.M. Lunch

1:00-5:00 P.M. I transmitted the eighth Coronary Artery Bypass Graft study's patient's ECG, and corrected all requested corrections by Abbott's monitor.

September 29, 2003

9:00 A.M. I arranged for the blood work to be collected from a patient in the Lap Chole study, and I scheduled the patient's appointment for tomorrow. I brought the patient's blood work to Quest Laboratories, filled out the requisition and labeled the tubes.

12:00 P.M. Lunch

1:00-5:00 P.M. I corrected all requested corrections by Abbott's monitor.

September 30, 2003

9:00 A.M. I went to class.

11:00 A.M. I corrected all requested corrections by Abbott's monitor.

I went to medical records to copy more of the Urokinase study's first patient's medical chart.

12:00 P.M. Lunch

1:00 P.M. The Coronary Artery Bypass Graft study's fifth patient arrived for the 30 day follow-up. We collected the patient's ECG, vitals, and blood work.

2:00 P.M. The Lap Chole patient has a follow-up appointment.

3:00 P.M.	I transmitted the fifth patient's ECG. We processed the fifth patient's blood work and shipped it to Covance via Federal Express.
4:30-5:00 P.M.	I continued correcting all requested corrections by Abbott's monitor.

October 2, 2003

9:00 A.M.	I went to class for review.
11:00 A.M.	I corrected all requested corrections by Abbott's monitor.
12:00 P.M.	Lunch
1:00 P.M.	I continued correcting all requested corrections by Abbott's monitor.

October 3, 2003

9:00 A.M.	I corrected all requested corrections by Abbott's monitor.
12:00 P.M.	Lunch
1:00-5:00 P.M.	I continued correcting all requested corrections by Abbott's monitor.

October 6, 2003

9:00 A.M.	I went to take my 5600 test.
11:30 A.M.	We went to OMCT to screen a candidate for the Urokinase study. The patient declined participation.
1:00 P.M.	Lunch

2:00-5:00 P.M.	I continued correcting all requested corrections by Abbott's monitor.

October 7, 2003

9:00 A.M.	I went to class.
11:00 A.M.	I corrected all requested corrections by Abbott's monitor.
12:00 P.M.	Lunch
1:00-5:00 P.M.	I continued correcting all requested corrections by Abbott's monitor.

October 8, 2003

9:00 A.M.	I corrected all requested corrections by Abbott's monitor.
12:00 P.M.	Lunch
1:00 P.M.	I continued correcting all requested corrections by Abbott's monitor.

October 9, 2003

9:00 A.M.	I went to class.
11:00 A.M.	I started working on my thesis and defense and calculated Urokinase administration values for the Urokinase study's second and third patient.
12:00 P.M.	Lunch
1:00-5:00 P.M.	I worked on my thesis and defense.

October 13, 2003

9:00 A.M.	I worked on my thesis and defense.
12:00 P.M.	Lunch
1:00 P.M.	I worked on my thesis and defense.

October 14, 2003

9:00 A.M.	I went to class.
11:00 A.M.	I worked on my thesis and defense.
12:00 P.M.	Lunch
1:00 P.M.	I worked on my thesis and defense.

October 15, 2003

9:00 A.M.	I worked on my thesis and defense.
12:00 P.M.	Lunch/Grand Rounds
1:00-5:00 P.M.	I worked on my thesis and defense, and I faxed Randy the specimen inventory.

October 17, 2003

9:00 A.M.	I went to the hospital to screen a Coronary Artery Bypass Graft patient. The patient agreed to participate and signed the informed consent.
12:00 P.M.	Lunch
1:00 P.M.	I transmitted the patient's ECG to Medifacts.
2:00 P.M.	I had a phone interview with MedTrials.
3:30-5:00 P.M.	I worked on my thesis and defense.

October 20, 2003

9:00 A.M.	I worked on my thesis and defense.
10:00 A.M.	I went to the hospital to screen a Coronary Artery Bypass Graft patient. The patient agreed to participate and signed the informed consent.
12:00 P.M.	Lunch
1:00 P.M.	I transmitted the patient's ECG to Medifacts
1:30 P.M.	I worked on my thesis and defense.
5:00-5:30 P.M.	I randomized a Linezolid patient, the first patient.

October 21, 2003

9:00 A.M.	I went to class.
11:00 A.M.	I received the news that the ninth CABG patient screen failed due to elevated creatinine levels.
12:00 P.M.	Lunch with Paul from Aerotek
1:00-5:00 P.M.	Worked on thesis and defense.

October 22, 2003

Worked on thesis.

October 23, 2003

9:00 A.M.	I went to class.
11:00 A.M.	I went on an interview with Aerotek.
2:00-5:00 P.M.	I worked on my thesis and defense.

October 24, 2003

9:00 A.M.	I went to Dr. Reeves', Dr. Anderson's, and Dr. Peska's office to get them to sign my intent to defend form.
11:00 A.M.	I went to Kindred Hospital and had blood collected for the Linezolid study's first patient.
12:00 P.M.	Lunch
1:00-5:00 P.M.	I worked on my thesis and defense.

October 27, 2003

9:00 A.M.	I worked on my thesis and defense and studied for my exam in 5600.
12:00 P.M.	Lunch
1:00-5:00 P.M.	I worked on my thesis and defense.

October 28, 2003

8:00 A.M.	There was a potential candidate for the Coronary Artery Bypass Graft study to inform, and the patient's pre-op was at 8:00 AM. The patient refused to participate in the study.
9:00 A.M.	I went to class.
11:00 A.M.	I got Dr. Peska to sign my Intent to Defend Form.
12:00 P.M.	Lunch
1:00-5:00 P.M.	I worked on my thesis and defense.

October 29, 2003

9:00 A.M.	I worked on my thesis and defense and studied for 5600 exam.
12:00 P.M.	Lunch
1:00-5:00 P.M.	I worked on my thesis and defense and studied for 5600 exam.

October 30, 2003

9:00 A.M.	I went to class
11:00 A.M.	I studied for my 5600 exam
12:00 P.M.	Lunch
1:00-5:00 P.M.	I worked on my thesis and defense and studied for 5600 exam.

October 31, 2003

9:00 A.M.	I worked on my thesis and defense and studied for 5600 exam. We had scheduled a potential CABG candidate. The patient did not qualify.
12:00 P.M.	I went to the department of surgery's floor Halloween party.
1:00-5:00 P.M.	I worked on my thesis and defense and studied for 5600 exam.

SIGNIFICANCE AND THE SPECIFIC AIM OF THE STUDY

Significance

FDA has informed Abbott Laboratories of additional concerns related to manufacturing deficiencies for urokinase (Abbokinase). Until these problems are corrected, further distribution of Abbokinase would violate federal laws designed to assure the safety of drugs for patient use. FDA's concerns about the product relate to serious deficiencies in the manufacturing processes, the testing of the product, and the screening and testing of the donors of the kidney cells used to make Abbokinase.

Abbokinase is derived from cultures of human kidney cells from newborns who have died of natural causes, and is approved in the United States to dissolve blood clots in the lungs and heart arteries. It is also approved to help clear intravenous catheters.

During inspections of Abbott Laboratories and of BioWittaker, Inc., Abbott's supplier of human kidney cells, FDA identified numerous significant deviations from current good manufacturing practice (CGMP) regulations designed to assure product safety.

Compliance with CGMP is important because products manufactured from human sources have the potential to transmit infectious agents. CGMP for products such as Abbokinase requires important, overlapping safeguards in the production process, including adequate

- screening of donors and testing of the cells,

- controls for proper harvesting, storage, and handling of materials used in all stages of manufacturing, and

- processes to remove or inactivate infectious agents from the product.

Over the past several months, the firm has reported to FDA that a number of in-process lots of Abbokinase were contaminated with microorganisms. Six such lots were found to contain various strains of reovirus, a virus that usually results in no symptoms or causes minor respiratory or gastrointestinal symptoms. Association of reovirus infection with other human diseases has been reported, although a causal link has not been established. Another in-process lot was contaminated with mycoplasma, a microorganism that can cause respiratory infections, and, on rare occasions, other infections that may be serious. Abbott has assured FDA that none of these in-process lots were manufactured into final product or distributed.

These recent findings of contamination and Abbott's inability to locate the source of the problem have raised further concerns at FDA about Abbott's entire manufacturing process for Abbokinase. Abbott's deviations from CGMP could significantly impact the safety of the product. One FDA concern is that deficiencies in manufacturing practices could also lead to the product being contaminated with microorganisms that have not yet been detected.

FDA also obtained additional information regarding the inadequacy of the screening and testing of the mothers and donors of the human kidney cells used to produce

Abbokinase. Information was also obtained regarding the seven instances of in-process lots of product being contaminated with reovirus and mycoplasma.

In the letter to Abbott, the agency has detailed the steps Abbott needs to take to correct the serious and significant manufacturing deviations. These include:

- completing a thorough and adequate investigation of the reovirus and mycoplasma contamination, including the source of the contamination,
- manufacturing Abbokinase using human kidney cells that have been obtained, processed, and tested through adequate methods, and
- assuring that fully validated methods are used in the manufacturing process to test for infectious agents and remove them.

Abbott submitted a supplemental new drug application providing for changes in procurement and processing of neonatal kidney cells, improvements in the manufacture and testing of the drug substance and drug product, revised release specifications for the drug substance and drug product, a revised CBER lot release protocol, withdrawal of the "Open-Cath" dosage strengths, and revised labeling. Labeling revisions include updated information regarding product source and adverse reactions, as well as withdrawal of the coronary artery thrombosis and catheter clearance indication.

The Department of Health and Human Services completed the review of that supplemental application, as amended, and it was approved based on Abbott's written commitments, one of which is

- To conduct a study to assess the immunogenicity of Urokinase after primary dosing.

Urokinase is indicated in adults for the lysis of acute massive pulmonary emboli, defined as obstruction of blood flow to a lobe or multiple segments, and for the lysis of pulmonary emboli accompanied by unstable hemodynamics, i.e., failure to maintain blood pressure without supportive measures. Therefore, it is important to complete this study, to meet federal requirements, so that Urokinase could be fully marketed and help to improve the quality of life.

This information in the Significance section has been obtained from articles on U.S. Food and Drug Administration website: http://www.fda.gob/cder/biologics/infosheets/abb032299.htm

Specific Aim

1.) To access the human antibody response to Urokinase, a thrombolytic agent in subjects treated for acute lower extremity ischemia.

a.) Technique:

All subjects will receive an intra-arterial infusion of a minimum of 240,000 IU of Urokinase (UK). In part A of this study we will obtain serum specimens from subjects receiving UK. These blood specimens will be used in part B of this study for the qualitative/quantitative assessment of antibody response to Urokinase, specifically IgM, IgE, IgG. Antibody directed against the UK drug substance, API, and the inactive peptides/protein in the formulation.

DISCUSSION

My internship at the University of North Texas Health Science Center involved training and hands-on activities associated with the clinical research process in the Department of Surgery.

I was under the guidance and direction of Della Weis, R.N., CCRC from 9:00 a.m. to 5:00 p.m., five days a week. I have developed skills that will enable me to:

1.) Coordinate a study according to the protocol and IRB regulations, using Good Clinical Practice (GCP) and Standard Operating Procedure (SOP) guidelines

2.) Understand the logistics of initiation, monitoring, and closeout visits

3.) Complete, file and understand all paperwork needed for a study (e.g. Financial disclosure, Serious Adverse Events (SAE) and Adverse Event (AE) reporting, Case Report Form (CRF) reporting, Source Documents, informed consent, etc…)

4.) Understand clinical literature about specific therapeutic area (e.g. Coronary Artery Bypass Graft, Lower extremity ischemia, intra-abdominal abscess)

5.) Become experienced in conducting informed consents, in-service meetings for staff, patient recruitment, resolving queries, and processing lab work

6.) Independently coordinate studies

7.) Understand the duties and responsibilities of the monitor, and as a coordinator, self monitoring trials

Throughout my internship experience in the Department of Surgery, I have co-coordinated four different studies.

1. Comparison Study of the Efficacy and Safety of Tigecycline to Imipenem/Cilastatin to Treat Complicated Intra-Abdominal Infections in Hospitalized Subjects, phase III
2. Linezolid vs. Vancomycin/Oxacillin/Dicloxacillin in the Treatment of Catheter-Related Gram-Positive Bloodstream Infections, phase IV
3. Assessment of Human Antibody Response to Urokinase in Subjects Treated for Acute Lower-Extremity Ischemia, phase IV
4. Study of the Safety and Efficacy of Parecoxib/Valdecoxib and Placebo/Valdecoxib Compared to Placebo for Treatment of Post-Surgical Pain in Patients who have Coronary Artery Bypass Graft via Median Sternotomy, phase III

In the United States, GCPs (Good Clinical Practices) are defined by federal law in the Code of Federal Regulations Title 21 and Title 45 and are enforced by the FDA. The purpose of GCPs is to protect the rights of subjects to privacy, autonomy, beneficence, safety, and to protect the integrity of clinical data. The clinical research process is strongly dependent on these exact GCPs.

Clinical investigation of an investigational drug must be conducted under specific clinical research regulations enforced by FDA. The clinical investigation is

generally divided into four phases. Phase 1 is the initial introduction of an investigational new drug into humans. Phase 1 trials are generally closely monitored and conducted in normal healthy volunteers. The focus of Phase 1 studies is to determine the drug pharmacokinetic and pharmacological effects. Phase 2 trials are conducted to evaluate the effectiveness of the drug for a specific indication and to determine common short-term side effects associated with the drug. Phase 3 trials are expanded controlled and non-controlled trials including several hundred to several thousand subjects. Phase 3 trials are conducted to gather additional information of effectiveness and safety to determine the overall benefit-risk relationship of the drug and to provide an adequate basis for the physician labeling. The Phases of Clinical Research can be found in the Code of Federal Regulations (21 CFR 312.21).

Phase 1 Clinical Trials

Purpose

Studies a drug's safety profile. This safety profile includes how a drug is absorbed, distributed, metabolized, excreted and duration of action.

Test Population

- Subjects are normal healthy volunteers or patients with terminal illnesses who have failed other therapy
- Sample size usually < 100 subjects

Phase 2 Clinical Trials

Purpose

Small, well-controlled trials to evaluate drug safety and efficacy; assess side effects; also referred to as dose ranging studies

Test Population

- Subjects are patients who have the disease/condition
- Sample size usually 100-300 subjects

Phase 3 Clinical Trials

Purpose

Verifies effectiveness, evaluates general benefit-risk assessment, monitors adverse reactions from long-term use.

Test Population

- Well-controlled, double-blind
- Sample size usually > 1,000 subjects

Phase 4 Clinical Trials

Purpose

- Postmarketing studies
- Observation by design
- Evaluates the drug's safety during routine use
- Identifies additional safety information

Clinical research activities can be subdivided into three basic categories including pre-study, active, and a post-study. Once the need for a clinical trial is established, pre-study activities begin. Pre-study activities continue until the initiation of the study. The active stage of the clinical trial involves heavily regulated testing of the drug or device with the study's chosen subject population. After completion of the clinical trials, post-study activities commence. The data are compiled, analyzed, and used to gain product approval by the Food & Drug Administration (FDA) or to make additional labeling claims about the product.

PRE-STUDY ACTIVITIES

Pre-study activities are central to developing a sound clinical investigation. The following table describes the major pre-study activities performed in Clinical Trials.

Table One: Pre-Study Activities[44]

Activity	Description of Activity	FDA CFR (If Applicable)
Literature Review	A review of current literature on the area of interest to aid in the development of the study protocol	
Protocol Development	The design by which the clinical study is to be conducted. Protocols are required for each study. Protocols include an objective statement, the study design along with any tests or procedures, study timetable, subject sample design, participant inclusion/exclusion criteria, participant exit criteria test article description and instructions for use, safety information, guidelines for data collection and adverse event reporting, and methods to prevent bias.	21CFR 312.23(6), 21CFR 812.25 (b)
Source Document Development	Forms that capture original study data from a patient's visit. These documents are usually a part of the patient chart maintained by the principle investigator (PI). Accurate source documents are the responsibility of the PI.	21 CFR 312.62
Case Report Form (CRF) Development	Forms used to collect study data. The information captured on the source documents by the investigative site is transferred to the case report forms for database entry and analysis by the sponsor.	21 CFR 12.62

Activity	Description of Activity	FDA CFR (If Applicable)
Institutional Review Board (IRB) Approvals	Institutional Review Boards examine certain documents pertaining to the use of human subjects for clinical trials. The IRB has the option of approving, disapproving, or requiring modifications to clinical documents, including the informed consent form and protocol. IRB review and approvals are designed to protect the study participant. The PI is responsible for ensuring that the IRB complies with part 56 of the CFR. The PI must update the IRB on any changes that occur in the study, unanticipated events and any deviations from the approved study protocol.	21CFR 56, 21CFR 312.66, 21 CFR 812.42, 21 CFR 812.60, 21 CFR 812.62, 21 CFR 812.64, 21 CFR 812.66
Principle Investigator (PI) Selection	The individual who actually conducts the trial, i.e., under whose immediate direction the test article is administered to the study participant.	21CFR 312.53, 21 CFR 812.3 (i), 21 CFR 812.43
Informed Consent Form Development	A legal document representing that the subject has had the study explained to them, understands the study, and voluntarily agrees to participate. The PI is responsible for obtaining informed consent from each study subject.	21 CFR 50.25
Study Budget/Principle Investigator Grants/ Financial Disclosure	The study budget is an estimate of all anticipated expenses involved in completing a clinical trial. The principle investigator is responsible for providing the sponsor with complete and accurate financial information, which allows the sponsor to submit and comply with the federal financial disclosure requirements in the referenced regulation.	21 CFR 54, 21 CFR 312.53, 21 CFR 812.43

Activity	Description of Activity	FDA CFR (If Applicable)
Pre-Study Visits	The pre-study visit is conducted by the sponsor and intended to evaluate the potential investigator, the investigator's staff, the investigator's equipment, the patient population, and the site. All aspects of the current study including target population, protocol design, and case report form completion are discussed.	
Ordering & Shipping Clinical Supplies	Clinical supplies may be either the test article or any supplies required to carry out the administration of the protocol	21CFR 52.108, 21 CFR 312.61, 21 CFR 312.62

Table Two: Active Study[44]

Activity	Description of Activity	FDA CFR (If Applicable)
Review Study Status	The monitor should keep up to date regarding general status of the trial. Assessment of study progress, patient enrollment, protocol compliance, adverse events, any changes in study participants or study staff are all ongoing steps to evaluate the study status.	21 CFR 312.56, 21 CFR 812.46
Inspect Study Binder	The monitor must inspect the study binder to make certain that all required regulatory documentation is accurate and up to date. Items to review include the signed Form 1572, curriculum vitae of investigators and subinvestigators, clinical investigator's brochure, protocol and amendments, IRB records, approved informed consent form, monitoring records, test article accountability records, correspondence, and telephone records.	21 CFR 312.56, 21 CFR 812.140
Review Informed Consent Forms	Each study subject should have signed an informed consent form. The monitor must examine the informed consent forms to make certain that the IRB approved version was executed and that all required signatures and dates are present	21 CFR 50
Review Case Report Forms	The monitor should review the case report forms making sure that the forms are current and accurate. A comparison between the case report forms and the source documents should be made to verify accurate transcription. The monitor oversees any corrections to the case report forms.	21 CFR 312.64
Review Clinical Supplies and Records	The records and storage of the test article must be inspected to confirm compliance with protocol. An inventory of clinical supplies should be made by the monitor to ensure proper documentation of receipt and dispensing to patients and avoid clinical supply emergencies.	21 CFR 312.61, 21 CFR 312.62, 21 CFR 812.140
Complete Monitoring Report	After assessing all of the areas above, a monitoring report should be completed summarizing all of the findings from the visit.	

Activity	Description of Activity	FDA CFR (If Applicable)
Discuss Results with PI and Staff	The findings of the monitoring report and the study status should be discussed with the PI and study staff. The monitor must explain any corrective actions required and field any questions from the PI or study staff.	
Obtain Review and Signatures of Study Manager and Medical Monitor	The final step in the monitoring visit involves the review and approval of the Study Manager and the Medical Monitor. Both of these individuals should review the monitoring report for possible violations and validate the study status with their signatures.	

Table Three: Close-Out Activities[44]

Activity	Description of Activity	FDA CFR (If Applicable)
Inspect Study Binder	The close-out visit is the final onsite inspection of the study binder. The binder must be inspected to make certain that all required regulatory documentation is accurate and complete. Items to review include the signed Form 1572, curriculum vitae of investigators and subinvestigators, clinical investigator's brochure, protocol and amendments, IRB records, approved informed consent form , monitoring records, test article accountability records, correspondence, and telephone records.	21 CFR 312.56, 21 CFR 812.140
Review Informed Consent Forms	This review is the final onsite inspection of the informed consent forms. Each study subject should have signed and dated an informed consent form. The monitor must examine the informed consent forms to make certain that all required signatures and dates are present.	21 CFR 50
Review Case Report Forms	The monitor should make a final, onsite review of the case report forms making sure that the forms are kept current and accurate. A comparison between the case report forms and the source documents should be made to verify accurate transcription. The monitor oversees any corrections to the case report forms.	21 CFR312.64, 21 CFR 812.140
Final Disposition of Test Article and Clinical Supplies	An inventory of test articles and clinical supplies must be checked against dispensation logs to account for all of the test articles and clinical supplies shipped to the site. The balance of test article must be returned to the sponsor. A Return and Disposal of Clinical Test Articles form must be completed, signed by the PI, and returned along with the supplies	21 CFR 312.59, 21 CFR 312.61, 21 CFR 312.62, 21 CFR 812.140
Discuss Results and Responsibilities with PI and Staff	The results of the monitoring visit are discussed with the PI and staff as well as record retention requirements. The investigator must submit a final report to the IRB within three months of the study completion.	21 CFR 312.62 (c), 21 CFR 812.140

Activity	Description of Activity	FDA CFR (If Applicable)
Lock of Study Database	After a thorough review of the data by the monitor, Biostatistics, and Product Safety, and after corrections are made to the database, the study data is locked so that no more modifications can take place.	
Preparation of Final Clinical Study Reports (CSR)	The Clinical Study Report (CSR) summarizes the clinical study. The CSR includes information on the purpose of the study, clinical procedures used to conduct the trial, and the statistical and clinical conclusions. The CSR is cooperatively prepared by the Study Manager, Biostatistician, and Investigational Product Safety. Information is obtained from other individuals or departments such as the Medical Monitor, Regulatory Affairs, Pharmacokinetics and Health Economics, as necessary. The final CSR must be approved by the Study Manager, Unit Head, Medical Monitor, Investigational Product Safety, Biostatistics, and Regulatory Affairs.	21 CFR 50, 21 CFR 54, 21 CFR 56, 21 CFR 312, 21 CFR 812

CLINICAL TRIAL MONITORING

Pre-Study/Qualification

Prior to conducting clinical trials, sponsors are obligated to select qualified investigators. The investigators must have the appropriate training and experience to perform the protocol-required assessments and evaluations in addition to having adequate staffing and facilities. The purpose of a pre-study visit is to ensure that qualified investigators are selected to participate in the clinical trial.

Pre-study visits are required by ICH guidelines and further specified in sponsor SOPs and FDA monitoring guidelines. The manner in which pre-study visits are conducted is highly dependent on sponsor policy and whether or not the sponsor has previously worked with the investigator at a given research facility.

Pre-study visits are usually conducted on-site at the investigator's office so that the sponsor has an opportunity to personally meet the research site personnel and to tour the facility where the research will be conducted.

For the Urokinase study, an actual on site pre-study visit was not conducted by the sponsor (Abbott Laboratories). Abbott Laboratories had previously worked with UNTHSC's site in the TOPAS study. Therefore, Abbott was comfortable with conducting the pre-study visit over the phone.

Many sponsors opt to conduct pre-study visit telephone interviews with investigators with whom they are currently conducting a study or who have successfully completed studies in the recent past.

For the Urokinase study, the pre-study telephone interview consisted of overviews of study and objectives, investigator obligations, sponsor obligations, budgeting, discussion of the protocol, questions and answers.

Initiation

The initiation visit is crucial to the success of the clinical trial. The purpose of this visit is to clarify the applicable regulations and requirements of the protocol and carefully review the actual process of implementing the protocol at the site. In the event that an investigator meeting has already taken place, the initiation serves as a reminder regarding study and sponsor-specific procedures. If no investigator meeting has been conducted, then the on-site initiation is critical in establishing both the intent and the tone of the relationship between site and sponsor.

The sponsor representative will assure that the investigator and research personnel understand and accept their obligations in conducting the clinical investigation and will conduct an extensive and in depth review of protocol-related topics.

For the Urokinase study, Della and I and Dr. Yurvati (Sub-Investigator) attended the investigator's meeting in Chicago. At the investigator's meeting our site learned more about the Urokinase protocol, CRFs, Safety Reporting, and

Regulatory compliance. Also, there were changes to the CRFs that were mentioned at this investigator's meeting; moreover, there were questions answered that the sites had about the protocol. Leaving the investigator's meeting, we completely understood what Abbott Laboratories was expecting from our site for this study.

There was an initiation visit two weeks after the investigator's meeting. Prior to this initiation visit, Della and I worked hard to ensure that the informed consent was approved through our IRB, that the Regulatory document files were complete, and that the CRFs were properly stored. Two of Abbott's representatives conducted the initiation visit of which Della and I were present. Dr. Peska (Principle Investigator) briefly attended the meeting. At this initiation visit the Abbott representatives briefly reviewed the study and objectives, being that we had just came from the investigator's meeting, reviewed the investigator obligations, reviewed sponsor obligations, and reviewed regulatory documents. Dr. Peska signed a delegation sheet revealing that he delegated certain authorities to Della and me. Then, we toured the pharmacy to ensure that our site received the drugs and that they were being properly stored.

We also had initiation visits for the Tigecycline study, Linezolid study, and the CABG study. All of these studies were prepared for and conducted in the same manner; however, the material was significantly different.

Routine Monitoring

Sponsors are obligated to conduct monitoring of clinical investigations. The purpose of the monitoring visits is to ensure protocol and regulatory compliance. Sponsor representatives visit the site at appropriate intervals to verify that the study is being conducted properly and that the site, staffing, enrollment rate, record keeping, reporting, equipment and facilities remain adequate.

Monitoring is required by ICH guidelines and further specified in sponsor SOPs and FDA monitoring guidelines. The manner in which monitoring is conducted is highly dependent on sponsor policy and whether or not the sponsor has previously worked with the investigator at a given research facility.

Monitoring visits are conducted on-site at the investigator's office so that the sponsor has an opportunity to review actual study documentation/files and to inspect the facility where the research is conducted.

For the Urokinase study, our first routine monitoring visit was after we had enrolled the first patient, and the sponsor representative came to our site and reviewed the patient's CRF/Source Document, our site's regulatory binder, and our drug supply in the pharmacy. The second routine monitoring visit was after we had enrolled the third patient. The monitoring visit was somewhat long and tedious for the monitor due to the volume of information stored in the CRFs and Source Documents. When the monitor (Abbott representative) came, I gave him the regulatory binder, all the three completed CRFs, and the three patient's source

documents (copies of the patient's medical chart). The monitor then asked me to go through all of the CRFs with him to verify all of the information that I had transcribed. The monitor flagged the pages that needed changes. The monitor also requested that a few changes be made to the regulatory binder. This was a two day visit. After the monitor's visit, we sent off the frozen antibody specimens to Abbott. The monitor came back three weeks later to retrieve the white copies of the CRFs so that the Data Management Department at Abbott could start building the study database. At this routine visit, we corrected everything that the monitor requested to be changed.

In the CABG study, the monitor's routine visits were more frequently because it is a very difficult and detailed study.

There were also routine visits for the Tigecycline, and Linezolid studies.

The Routine monitoring for all four of the studies consisted of these seven duties:

1. **Staffing**
    - Have there been any changes in study personnel?
    - Is the staffing sufficient?
2. **Enrollment**
    - Is study enrollment lower than anticipated?
    - Are the screening/enrollment logs being completed and submitted on a timely basis?
3. **CRF Completion**

- Is CRF completion on schedule?
- Are outstanding CRFs complete and available for review?
- Does the site need any additional forms/supplies?

4. **Problems/Deviations**

- Are there any questions or concerns?
- Have there been any protocol deviations since the last monitoring visit?
- Are there any outstanding data queries?
- Are there any difficulties with processing/shipping laboratory specimens?

5. **Adverse Events**

- Have there been any serious adverse events (SAEs) since the last monitoring visit?
- Have all SAEs been reported to the sponsor and to the IRB?
- Have sponsor generated safety reports been submitted to the IRB?

6. **Regulatory Documents**

- Are there any updates? Obtain copies of correspondence, renewals and revised study-related documents

7. **Review of Subject Records**

During monitoring visit, the monitor should compare a representative number of subject records and other supporting documents with investigator's reports to determine that:

- The data recorded in the investigator's reports are complete, accurate and legible.
- There are no omissions in the reports of specific data elements such as the administration of concomitant test articles or the development of an intercurrent illness.
- Missing visits or examinations are noted in the reports.
- Subjects failing to complete the study and the reason for each failure are noted in the reports.
- Informed consent has been obtained and documented in accordance with good clinical practices.

I had an in-depth and very detailed experience with duties #3 and #5. I was responsible for completing the Urokinase CRFs. This work was very tedious and time consuming. First, I would go to OMCT Hospital and make copies of the patient's chart. Then I would sort the chart into sections (physician's notes, physician's orders, nurse's notes, nurse's flowcharts, medications). Then, I would read each page of the medical chart very carefully. Using the information that I read in the chart I would fill out the CRF while highlighting the section where I retrieved the information. All the while, I would be constantly keeping a patient narrative of everything that happened when the patient was in the hospital and throughout all of the patient's follow-up visits. I completed each CRF with the most efficiency because I knew that all CRFs would be reviewed for accuracy and completeness by Abbott's monitor during the routing monitoring visits.

The adverse event documentation was very important to this study. For the Urokinase study, we did come across AEs and SAEs. After being discharged from the hospital, Subject 1 required an Aortal Bi-femoral bypass. The monitor felt that the need for that surgery may be an SAE. I called the Medical Monitor and she stated that Abbott is FDA exempt for that aforementioned surgery and the need for the surgery should only be documented as an AE. The Medical Monitor contacted me later and stated that the need for the surgery is not even and AE, so there would be no subsequent paperwork to follow. That weekend, the patient was diagnosed with Compartment Syndrome which lead to the performance of a Fasciotomy. Inevitably, the need for this surgery was definitely a SAE. Then, I followed up with the subsequent paperwork.

First, I looked up the definition of an SAE for this study (See Appendix B). Since the Fasciotomy was indeed an SAE, I had to notify Abbott and fax the forms listed on the Cover Memo in Appendix C. Next, I had to notify our IRB and send them everything that I had sent to Abbott and our IRB onsite SAE application.

Subject 2, developed bilateral groin site hematomas which was considered an AE (See Appendix B). According to Abbott, "The investigator will assess and record any allergic, other non-hemorrhagic and hemorrhagic AE in detail on the specific AE CRF including the date of onset, description, final diagnosis, syndrome (if known), severity, time course, duration and outcome, relationship of

the AE to the study drug, and any actions taken." I filled out the subsequent paperwork and the monitor reviewed the information.

Study Close-out

At the conclusion of a clinical trial, sponsors are obligated to close out the participating investigational sites. The purpose of a closeout visit is to ensure that all required study activities are complete at a given investigational site such as:

- All data collected and verified
- Final accounting and disposition of the investigational product
- Final reconciliation of study supplies
- Study files are complete and correct

Closeout visits are typically conducted when all study visits are complete. Closeout visits are required by ICH guidelines and further specified in sponsor SOPs and FDA monitoring guidelines.

I had interactions with the closeout visit of the Randomized, Placebo-Controlled, Parallel Group Study to Evaluate the Safety and Efficacy of Ear Wax Removal Solution in Patients with Excessive or Impacted Cerumen in the External Auditory Canal. Basically, the monitors from Alcon, reviewed all CRFs and Source Documents and flagged the pages where they felt the information documented was inconsistent with the source documents. I helped Della answer all of the queries. Then the two Alcon monitors collected the empty boxes of the study supplies.

All records are kept and stored. An investigator is required by law to retain all study related records for "a period of two years following the date a marketing application is approved for the drug for the indication for which it is being investigated."

A GLANCE AT THE UROKINASE STUDY

The Urokinase study is a Phase 4, open-label, single-arm, multi-center clinical trial. All subjects enrolled received Urokinase. A total of 250 subjects will be enrolled at up to 50 investigational centers in order to guarantee complete blood sample sets from a minimum of 200 subjects.

Blood specimens were used for the qualitative/quantitative detection of antibodies to urokinase, specifically IgM, IgE, and IgG antibodies against:

- The drug substance
- The active pharmaceutical ingredient (API)
- The unbound fraction from the benzamidine-sepharose column

Della, Chris, and I were on the look out for potential candidates for this Urokinase study. We were looking for patients who were able and willing to give written informed consent, ≥18 years of age, with signs and symptoms of acute lower-extremity ischemia, arteriographically confirmed occlusion of the native artery or bypass graft located below the aortic bifurcation that is presumed to be clot. We were not looking for patients who had recent (within 10 days) major surgery, obstetrical delivery, organ biopsy, previous puncture of non-compressible vessels, or serious gastrointestinal bleeding; patients with a high likelihood of a left hear thrombus, subacute bacterial endocarditis, hemostatic defects including those secondary to severe hepatic or renal disease; cerebrovascular disease; diabetic hemorrhagic retinopathy; or any

other condition in which bleeding might constitute a significant hazard or be particularly difficult to manage because of its location; contraindication to heparin; allergy to contrast agents that the investigator feels cannot be properly premedicated; pregnant or nursing; life expectancy of less than one month; any known immunodeficiency disease or disorder, e.g., HIV; receiving immunosuppressant therapy for any reason.

Since, we had the population of patients of interest narrowed down, now, we had to come up with a strategy to recruit these patient that met these aforementioned inclusion/exclusion criteria. Dr. Peska (PI), Dr. Yurvati (Sub-Inv.), and Dr. DeLange (Sub-Inv) were our initial target being that they are all vascular surgeons who were involved in the study. So, we used the referable candidates from our three surgeons on the Urokinase study.

For the first patient, Dr. Peska informed us that there was a potential Urokinase candidate and the patient was pre-oping on a certain day. An hour before the patient was to enter surgery, Chris and I introduced ourselves to the candidate and family and explained the study to the candidate and family. The patient agreed to participate, then, we obtained the informed consent, having the patient initial and date every page and sign the last page (See Appendix A). Next, we performed pretreatment assessments.

- Demographic (age, gender, race)
- Stated height and weight
- Recent (within 6 months) medical history

- Peripheral vascular disease history
- Tobacco and alcohol history
- Medication history
- Vital signs (blood pressure, temperature, pulse rate)

The routine laboratory specimens required for the study were obtained prior to our arrival. These routine laboratory specimens were as follows:

- WBC, hemoglobin, hematocrit, platelet count
- aPTT
- Pregnancy test (if applicable)

Next, we obtained 4 red top tubes for the pre-treatment antibody assessment blood sample. Now, the patient was officially considered enrolled in the study.

As the patient was getting prepped, Chris, Dr. Peska, and I went to the OR (Operating Room). Dr. Peska wrote an order to have the pharmacy prepare the urokinase. We then brought the order to the pharmacy and brought the urokinase back to the OR. Then, Dr. Peska injected the urokinase as a bolus.

Then we went to the lab and centrifuged and aliquoted the specimens and stored the specimens to the -70 freezer.

Then, I would go back to the Patient Care Center and complete my narrative of the first patient's participation activity. Then, I would transcribe as much of the collected history and medical charts into the CRFs as I could while closely observing for any AE, SAEs.

After release from the hospital, the patient was asked to come back for the post-treatment procedures. We would collect additional blood specimens for antibody testing at three days, two weeks, and one month. Then we would continue to collect all serious adverse events for 30 days. We carried out these aforementioned series of event using these Schedule of Assessments listed below.

Schedule of Assessments

	Pretreatment	Treatment Period	3 days	7 days	2 weeks	1 month
Eligibility Criteria	X					
Informed Consent	X					
Medical History [a]	X					
Current Medications [b]	X					
Vital Signs	X					
Blood Collection [c,d]	X		X		X	X
Pregnancy Test (if applicable)	X					
Angiogram	X	X [e]				
Study Medication [f]		X				
Assessments of Adverse Events [g]	X	X	X	X	X	X

[a] General medical history (over last 6 months), PVD history, tobacco and alcohol history, demographics.

[b] Medications administered within 24 hours prior to enrollment, during treatment, and for 48 hours following end of treatment.

[c] Labs pretreatment include antibody testing, CBC and aPTT.

[d] Required post-treatment labs include antibody testing only.

[e] Performed per hospital protocol.

[f] Minimum infusion of 240,000 IU. Continued at Investigator's discretion.

[g] All AEs collected for 7 days or hospital discharge, whichever is sooner; SAEs collected for 30 days.

This information was obtained from Abbott Laboratories' manufactured CRFs.

For Subject 2 and 3 the procedures were the same; however, the drug administration was somewhat different. This was very tricky because Subject 1 was administered Urokinase as a bolus; however, Subject 2 and 3 received Urokinase administration over a drip. For the Treatment section of the CRF the Study Drug administration for a bolus is simple. The start date and end date is the same and the start time and end time is the amount of time it took to administer all of the bolus from the syringe. However, for a drip, the start dates, end dates, start times, and end times tend to fluctuate because drug administration also occurs on the floor. Usually for the angiogram the surgeon will request that Urokinase be administered in so many cc's at a given rate and when the patient reaches the floor the amount of drug being administered and the rate may change. I had to figure out how to convert cc/hr into IU/min for each change in rate. For example, let's say that the surgeon requested 1,200,000 units of Urokinase in 500 cc of Normal Saline at a rate of 70 cc/hr, I would calculate the following:

1,200,000 units/500 ml= 2400 IU/ml 70 ml/60 min=1.17 ml/min

then

2400 IU/ml X 1.17ml/min=2808 IU/Min

I feel as if I had the most wonderful and involved experience in the Department of Surgery. I have had a broad exposure to the pharmaceutical and medical device industry.

The clinical research industry is very thorough and attempts to prove safety and efficacy for public use of drugs and devices. I have learned that a Clinical Research Coordinator is the backbone of a study at the site level. The CRC is responsible for knowing the rules and regulations while being able to adhere to the patients, principal investigators, and the sponsor. My exposure to the industry left the impression that clinical research professionals have extreme responsibilities that they take very seriously. Della, Chris and I have had experiences where we would have to be at the hospital at 6 AM or 9 PM in order to randomized or inform a patient. I have learned that being a CRC is a very demanding and underpaid profession. There is a lot of hard work, time, and consideration that goes into this profession. Also, there is no room for mistakes because according to the FDA ignorance is not an excuse; therefore a CRC has to be really familiar with all rules, regulation, and protocols of FDA, ICH, IRB, and the sponsor. While most of my internship was based on the role of a Clinical Research Coordinator, I have come to respect and understand the primary roles of the other departments. It is now evident how all the other departments fit into the puzzle. For example, I have had exposure with the Regulatory Coordinator and the Grant Coordinator and I can see how they are critical to the smooth sailing of a study. For the CABG study, the monitor from Pfizer, Della and I had to look over the informed consent and the monitor informed us of what Pfizer would not allow in the informed consent and we took the revisions to the Regulatory Coordinator and she informed us of what our IRB would not allow in the informed consent, and then we came up with a compromise. Also, from the clinical trials aspect, I can see the importance of drug development. Now, I can understand why

there are so many side effects on the side of a Tylenol bottle. The effort of a clinical site to accurately follow the protocol, thoroughly explain an informed consent, to work according to GCP, and a score of other cooperative efforts to further the common goal of developing safe and effective pharmaceuticals is the greatest impression gained from my internship experience in the Department of Surgery at UNTHSC.

Appendix A

Informed Consent

Date

Protocol # Insert the Protocol #

INFORMED CONSENT AUTHORIZATION TO PARTICIPATE IN A RESEARCH STUDY

TITLE: **Insert the Name of the Study**

SPONSOR: **Insert the Name of the Sponsor**

INSTITUTION: **Insert the Name of the Institution**

PRINCIPAL INVESTIGATOR: **Insert the Investigator's Name**

SUBJECT NAME (Please Print):

Before agreeing to take part in this study, it is important to carefully read the following explanation of the intended procedures. This consent form may contain words that you do not understand. Ask the study doctor or the study staff to explain any word or information that is not clear to you. Also, understand that if you decide not to take part in this study, your treatment will not be affected.

1. STUDY PURPOSE

Based on your symptoms and tests, you have been diagnosed as having a blockage in a major artery (or bypass graft) in your leg, and your doctor has recommended that urokinase (Abbokinase®) be used to treat your blockage. A bypass graft is an artificial passageway for blood. Most blockages are caused by a blood clot in the artery (or bypass graft) that supplies blood to your legs.

There are two initial methods to treat this problem. One method uses "thrombolytic agents" or clot dissolving drugs, to dissolve the clot within a blood vessel or graft, usually followed by surgery to correct the underlying problem. The other method does not use thrombolytic agents, but uses surgery alone as treatment. At present, it has not been proven that one method is better than the other in providing patient benefit. Currently, no thrombolytic agent is approved by the FDA for use in this manner. However, urokinase (Abbokinase®) has been used to clear similar

types of blood clots. Abbokinase® is currently approved by the FDA for the treatment of massive pulmonary embolism (blood clots in the lungs). The purpose of this research study is to determine if treatment with urokinase causes the body to develop a response that may cause drug resistance or allergic reactions. In order to determine this, some of your blood will be collected, frozen and tested in the future by the sponsor for those possible responses. While the use of urokinase for this indication is still experimental (not yet approved by the FDA), the procedures used in this study are not experimental and are considered standard of care.

II. STUDY PROCEDURES

About 250 subjects will participate in this study at about 50 different hospitals in the United States. All subjects will receive urokinase. Your total participation time in the study is about one (1) month. The study will take about eighteen (18) months to complete. If you choose to participate in this study:

1. You will have a medical history (including medication history) and vitals signs taken.
2. An angiogram (special type of x-ray for blood vessels) will be performed to see if you do have a blood clot in your leg.
3. After it is found that you have a blood clot in an artery or graft in your leg, you will have about one tablespoon (10 mL) of blood drawn for future antibody testing.
4. A long tube called a catheter will be inserted through the skin into a major artery in your leg and moved to the site of the blood clot. Urokinase will be delivered into the clot through the catheter in order to try to dissolve the clot. The dose of urokinase used and the length of the
   treatment will be decided by your doctor.
5. Another angiogram (x-ray) of your leg artery or graft may be done during and at the completion of your treatment as directed by your doctor.
6. Your doctor may stop treatment at any time without your consent if necessary for your safety.
7. You will have a second blood specimen taken at about 3 days following the initiation of your treatment with urokinase.
8. You will be watched for adverse events during your hospital stay.
9. You will be asked to stay in the study for about one month and to agree to return for additional blood sampling at two weeks and one month following the start of your treatment. The total amount of blood that will be drawn at each visit is about 10 mL (less than one tablespoon). If any blood draw is missed you should still return to have the remaining blood samples taken.

Time and Travel Expense Compensation

You will be responsible for the usual and customary costs associated with treatment of your disease. You will receive $XX.XX for each completed blood draw (total of 4 blood draws) required for the antibody testing as compensation for your time and travel expenses in this study. These blood draws are not required for the treatment of your condition.

III. RISKS AND DISCOMFORTS OF THE STUDY

Some of the risks involved with this study are:

1. The risk of bleeding is increased with the use of urokinase. Deaths due to hemorrhage (bleeding), including bleeding in the brain and intestinal cavity (space where intestines are located), have been reported.
2. Low blood pressure or allergic reactions have also been seen during administration of urokinase. Rare cases (less than 1%) of fatal allergic reactions have been reported.
3. Reactions to urokinase infusion have included: low oxygen in the blood, shortness of breath, increased heart rate, low blood pressure, high blood pressure, acidosis (increased acid levels in the blood), fever, shaking chills, back pain, vomiting, and nausea.
4. Other adverse reactions reported using Abbokinase® in clinical studies include heart attack, recurrent blood clot, paralysis (loss of ability to move muscles), stroke (sudden rupture or obstruction of an artery of the brain), chest pain, sweating, and changes in your red cell and platelet counts (platelets are small strictures in blood that help it to clot).
5. Additional reports of adverse events include cardiac arrest (stopping of the heart), vascular embolization (obstruction of a blood vessel) including cholesterol emboli (obstruction caused by blood fat), cerebral vascular accident (accident involving blood vessels in the brain), pulmonary edema (fluid in the lungs), and reperfusion ventricular arrhythmias (uneven heartbeat when blood flow is being restored to an organ). (Please discuss with your doctor if you do not understand any of these terms.)
6. Urokinase is made from human tissue. Products made from human material may contain known and unknown infectious agents, such as human viruses, that could possibly have escaped the sterilization process during manufacturing. However, no viral infections from urokinase have been reported.
7. Some of the blood clot could break loose during the procedure and block the artery further down your leg. This could threaten your limb, but usually responds to continued therapy.
8. The effects of urokinase on a pregnant female or an unborn child are not fully known. For this reason, if you are pregnant, think you may be pregnant, or are trying to become pregnant, you must not participate in this study. You will have a pregnancy test done if you are able to become pregnant. You must not participate in this study if the test shows you are pregnant. If you find out later that you were pregnant at the time of receiving the study drug, you agree to be observed by the study doctor at least once every three months throughout your pregnancy. You will allow your baby's progress to be monitored for the first year of his/her life.
9. It is not known if this drug gets into breast milk or its possible effect on the feeding infant. Therefore, if you are breast-feeding a child, you must not participate in this study.
10. There may be mild discomfort or bruising associated with the blood drawing and occasionally a person feels faint. Rarely, an infection may develop, which can be treated.
11. There may be other risks and discomforts that are not known and cannot be predicted.

IV. CONTACTS

If a study-related problem should occur, or you experience an adverse reaction, or if you have any questions at any time about the study, you may contact the Principal Investigator.

This study has been approved by the Institution's IRB, which is the committee given responsibility by the FDA to make sure that the rights of persons like yourself are protected. If you have any questions about your rights as a participant in this study, you may contact the chairman of the Institution's IRB.

V. BENEFITS

In many cases, thrombolytic therapy allows the treatment of the blood clot before surgery. Medical opinion suggests that this therapy may help provide a more accurate assessment of the problem and allow for better planning of surgery after dissolving some of the blood clot. Reopening the artery may make surgery easier and the results with the initial clot-dissolving drug may be better than with surgery alone. In some cases, removing the clot with the thrombolytic drug may be the only treatment needed. However, there are no guarantees or promises that you will receive any medical benefit from participating in this study.

VI. ALTERNATIVE TREATMENTS

No clot dissolving medication is currently approved by the FDA to be given directly into a clot in a leg artery. Four blood clot-dissolving medications; urokinase (Abbokinase®), tissue plasminogen activator (t-PA), reteplase (r-PA), and streptokinase (SK) have been used to dissolve clots in the leg. Other ways to treat the blockage in your artery include injection of SK into a vein, mechanical removal of clot by means of a special catheter, and surgery. You may also choose to have no treatment. After you have received your initial treatment, your doctor will advise you whether additional treatment is needed.

VII. CONFIDENTIALITY

The medical information collected from this study will be submitted to the sponsor, to the Food and Drug Administration of the United States Government, and possibly to other regulatory agencies. This will be done under the regulations issued by the federal agency. To do this research, we need to collect health information that identifies you. The information we might use or disclose includes: supporting information from your entire medical record, results of lab tests, x-rays or other images, information from follow-up visits. The information collected in this study will be processed to meet the purpose of the clinical study. Information may be used for seeking approval from medicines regulatory authorities to market the medicine. It may also be used in reports of the

study or for scientific presentations. The sponsor may also use the information from this study which relates to you for future research, development, regulatory and manufacturing activities related to Abbokinase®. The sponsor, the Institution's IRB, and possibly regulatory agency personnel will review your medical records. This is to verify the procedures and information collected by the doctor. This will be done without violating your confidentiality to the extent allowed by law. By signing this consent, you are also allowing your records to be reviewed. Your identification (name and address) as a participant in this study will be filed at your hospital under adequate security. The sponsor may need to contact you for future follow-up studies. You will be notified of any significant new findings in writing. If the results of this study are published, your identity will not be revealed.

In addition, the sponsor, its related companies and other companies hired by the sponsor will have access to your personal information to process, analyze, and store data as part of the study. This information will be provided in a way that does not identify you. The information collected as part of the study may be transferred to these organizations by computer.

You have the right to request a copy of your records and make corrections, but some data may need to be withheld until after the study data analysis is completed.

Your Protected Health Information may no longer be protected by HIPAA (Health Insurance Portability and Accountability Act) once it is disclosed by your doctor, although other confidentiality safeguards apply. The sponsor will take all reasonable steps to protect your right to privacy.

You may cancel this authorization at any time by notifying the study doctor in writing. Unless revoked (cancelled), this authorization will never expire.

You do not have to sign this form. If you decide not to sign this form, you cannot be in the research study.

VIII. COMPENSATION FOR INJURY

If you are injured as a direct result of the administration of urokinase, the sponsor will pay all medical expenses necessary to treat the injury.

Date Protocol# Insert the Protocol #

You should know that by signing this form, you are neither waiving any of your legal rights against, nor releasing the sponsor, the principal investigator, the Institution or any of their respective agents from liability for negligence with respect to the conduct of this study.

IX. LEAVING THE STUDY

1. You may choose not to participate in this study without loss of benefits or prejudice to your care.
2. You may withdraw from the study at any time without loss of benefits or prejudice to your care.
3. Withdrawal from the study does not automatically revoke the authorization to use or disclose your personal information; the request to revoke authorization to use or disclose your personal information must be received in writing. The request to revoke authorization does not include information that has already been disclosed or information gathered as a result of your participation in the study. Information given to the sponsor before you cancel your authorization may still be used by the sponsor. The sponsor may also reanalyze the results of the study at a later date and combine them with results of other studies.
4. You have the right to ask the doctor any question concerning this study at any time.

X. NEW FINDINGS

You will be informed of any new information that becomes known during the course of this study which might affect your willingness to continue participation.

Date Protocol # Insert the Protocol #

XI. CONSENT

I have read and understand the information describing the study. All my questions have been answered to my satisfaction and I am signing this consent form voluntarily to indicate my choice to participate in this study. I understand that I will receive a signed and dated copy of this form.

Study Volunteer Name (print) Signature Date

Legal Representative [If Applicable] (print) Signature Date

Relationship to Patient:

Person Conducting Informed Consent Discussion (print) Signature

Copy given to patient by (print): Signature

Appendix B

Glossary

Glossary

Adverse experience (AE): Any untoward change or worsening of a research subject's baseline health conditions regardless of severity or causality. The severity of an adverse experience is a clinical determination, while the seriousness of an adverse event is a regulatory determination.

Blinding: A procedure in which one or more parties during the conduct of a study are kept unaware of the treatment assignment(s).

CABG: Coronary Artery Bypass Graft

Case Report Form (CRF): A data collection tool used to report required information per the protocol to the sponsor for each subject in a trial. CRFs may be printed, optical or electronic documents.

Clinical Research Coordinator (CRC): Study coordinators assist principal investigators to implement research protocols at the clinical sites. A CRC coordinates protocol-required tests and procedures, completes required paperwork and acts as a liaison between the site and sponsor.

Clinical Trial: The systematic study of a test article (treatment, drug, or device) in one or more human subjects. Clinical trials are intended to discover or verify the clinical, pharmacological, and/or pharmacodynamic effects of an investigational product(s) and to identify any adverse reactions and or to study absorption, distribution, metabolism and excretion of an investigational product(s) with the goal of ascertaining its safety and or efficacy.

Code of Federal Regulation (CFR): Specifically, Title 21, good clinical practices (GCPs) defined by Federal Law and enforced by the FDA. Enforced to protect subject's rights to privacy, beneficence, autonomy and safety to ensure the integrity of data.

Comparative study: One in which the investigational product is compared against another product.

Exclusion Criteria: Criteria which exclude a potential subject from participation in a study.

FDA: The United States regulatory authority charged with protecting the health and safety of consumers by ensuring that drugs, biological, and medical-device products, foods, dietary supplements and cosmetics are safe and are manufactured under sanitary conditions. FDA is also responsible for ensuring that all product labeling is truthful, informative and not deceptive. FDA grants IND (investigational new drug) and NDA (new drug application) approvals.

Form 1572: A legal commitment by the principal investigator to comply with all federal regulations regarding the conduct of a clinical research trial. The signed original is kept in the regulatory binder.

Good Clinical Practices (GCPs): A standard for the design, conduct, performance, monitoring, auditing, recording, analysis and reporting of clinical trials that provides assurance that the data and reported results are credible and accurate and that the rights, integrity and confidentiality of trial subjects are protected.

Inclusion Criteria: Criteria that prospective subjects must meet to be eligible for participation in a study.

Informed Consent: Written confirmation of a subject's voluntary agreement to take part in a particular trial must be documented in accordance with the relevant regulations of the legal jurisdiction in which the trial takes place. Consent should be sought only after subjects are given complete information about the trial including explanation of its objectives, potential benefits, risks and inconveniences, and the subject's rights and responsibilities as described in the Declaration of Helsinki.

Initiation visit: A meeting conducted by the sponsor prior to the official start of an investigation to assure that the principal investigator understands the investigational status of the test article, the controls, the nature of the protocol or investigational plan and accepts his or her obligations in the conduct of clinical investigation as set forth in parts 21 CFR 312, 812 and other applicable regulations.

Institutional Review Board (IRB): Any board, committee or other group formally designated by an institution that reviews, approves initiation of and conducts periodic reviews of ongoing biomedical research involving humans (21 CFR 56.102). All clinical studies performed in the United States as well as those conducted in many other countries, require the approval by or notification of an IRB or similar body.

Investigator: The person(s) ultimately responsible for the conduct of a clinical trial. This includes the health and welfare of subjects during the investigation. An investigator is defined as the individual under whose immediate direction a test article is dispensed, administered or used which involves a study subject.

Monitor: An individual appointed by the sponsor to oversee the progress of a clinical investigation (21 CFR 52.3). A monitor is responsible to the sponsor for monitoring the reporting the progress of a clinical trial and for data verification.

New Drug Application (NDA): The application for a license to market an investigational drug may be submitted to the FDA after Phase-3 clinical trials are complete. After the NDA is approved, the sponsor may distribute the product for sale.

Open Study: A trial in which subjects and investigators know which product each subject is receiving.

Placebo: A pharmaceutical preparation that contains no active agent.

Postmarketing surveillance: Ongoing safety monitoring of marketed drugs.

Protocol. A document that describes the objective(s), design, methodology, statistical consideration and organization of a trial. The protocol usually also gives the background and rationale for the trial, but these could be provided in other protocol referenced documents.

Protocol Amendments: A written description of a change(s) or formal clarification to a study protocol.

Randomization: The process of assigning trial subjects to treatment or control groups using an element of chance to determine the assignments in order to reduce bias.

Serious Adverse Event (SAE): Any adverse drug experience that results in any of the following outcomes: death, a life-threatening event, inpatient hospitalization or prolongation of existing hospitalization, a persistent or significant disability/in capacity or a congenital anomaly/birth defect. Important medical events, as based upon appropriate medical judgment, that may not result in death, be life-threatening, or require hospitalization may be considered a serious adverse drug experience when they may jeopardize the patient or subject and may require medical or surgical intervention to prevent one of the outcomes listed above (21 CFR 312.32)

Source Documents: The original place where data are recorded is considered to be the source documents for that information. Source documents include; hospital records, clinic and office charts, laboratory notes, memoranda, subject's diaries, or evaluation checklists, pharmacy dispensing records, recorded data from automated instruments, certified copies or transcriptions, microfiches, photographic negatives, magnetic media, x-rays, and subject files or records kept in pharmacies, laboratories and medico-technical departments involved in the clinical trial.

Sponsor; An individual, company, institution, or organization which takes responsibility for the initiation, management, and/or financing of a clinical trial.

Standard Operating Procedure (SOP): Detailed, written instructions for the purpose of achieving uniformity of the performance of specific functions.

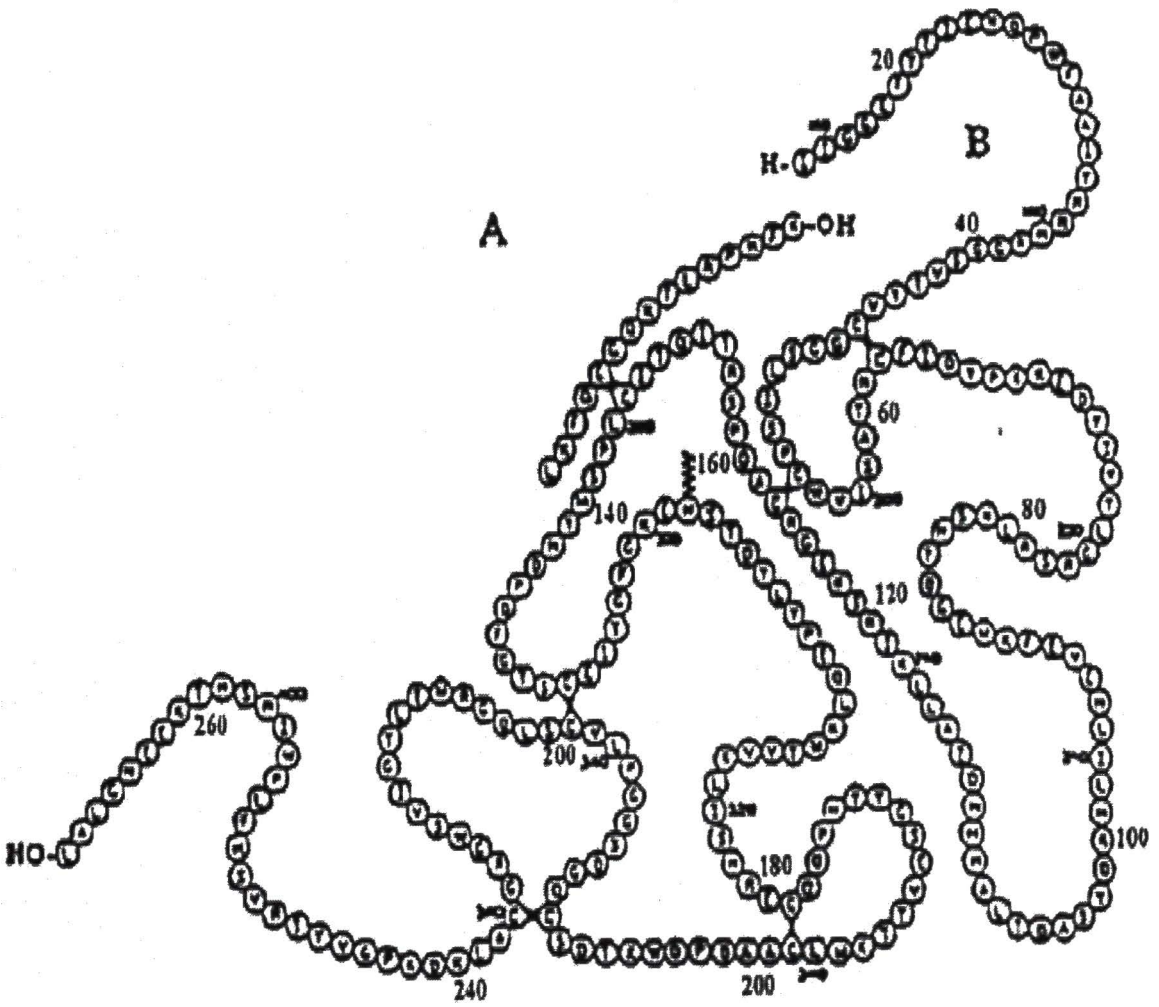
Sub-investigator: Any individual member of the clinical trial team designated and supervised by the investigator at a trial site to perform critical trial-related procedures and/or to make important trial related decisions.

These definitions were obtained from MedTrials, Inc., Introduction to Clinical Research and Studies.

Appendix C

Figure 1.1 (Urokinase)

The two-dimensional amino acid sequence of urokinase:
This pictorial representation was obtained from the Investigator's Brochure



REFERENCES

1. Urokinase-Pulmonary Embolism Trial Study Group, Urokinase pulmonary embolism trial, phase I results. JAMA 1970; 214: 2163-72.
2. Urokinase-Pulmonary Embolism Trial Study Group, Urokinase-streptokinase embolism trial, phase II results. JAMA 1974; 229: 1606-13.
3. A comparison of tissue culture urokinase and urinary source urokinase in patients with pulmonary embolism. Abbott Laboratories, 1976, Internal Scientific Report.
4. MacFarlane RG, Biggs R. Fibrinolysis. Its mechanism and significance. Blood 1948; 3: 1167-87.
5. Tillett WS, Garner RL. The fibrinolytic activity of hemolytic streptococci. J Exp Med 1933; 58: 485-502.
6. Milstone H. A factor in normal human blood which participates in streptococcal fibrinolysis. J Immunol 1941; 42: 109-16.
7. Christensen LR. Streptococcal fibrinolysis: a proteolytic reaction due to a serum enzyme activated by streptococcal fibrinolysis. J Gen Pysiol 1945; 28: 363-83.
8. Kaplan MH. Nature and role of the lytic factor in emolytic streptococcal fibrinolysis. Proc Soc Exp Biol Med 1944; 57: 40-3.

9. Tilett WS, Sherry S. The effect in subjects of streptococcal fibrinolysin (streptokinase) and streptococcal desoxyribonuclease on fibrinous, purulent, and sanguinous pleural exudations. J Clin Inv 1949; 28: 173-90.

10. MacFarlane RG, Pillin J. Fibrinolytic activity of normal urine. Nature 1947; 159: 779.

11. Astrup T, Sterndorff I. An activator of plasminogen in normal urine. Proc Soc Exp Biol Med 1952; 81: 675-8.

12. Sobel GW, Mohler SR, Jones NW, *et al.* Urokinase: an activator of plasma profibrinolysin extracted from urine. Am J Physiol 1952; 171: 768-9.

13. Lesuk A, Terminiello L, Traver JH. Crystaline human urokinase: some properties. Science 147: 880-2, 1965.

14. Bernik MB, Kwaan HC. Originof fibrinolytic activity in cultures of the human kidney. J Lab Clin Med 1967; 70: 650-61.

15. Bernik MB, Kwaan HC. Plasminogen activator activity in cultures from human tissues. An immunological and histochemical study. J Clin Invest 1969; 48: 1740-53.

16. Barlow GH, Lazer L. Characterization of the plasminogen activator isolated from human embryo kidney cells: comparison with urokinase. Thromb Res 1972; 1:2 01-6.

17. Weiss RE, Schleicher JB. A multisurface tissue propagator for the mass-scale growth of cell monolayers. Biotech and Bioengr 1968; 10: 601-15.

18. Weiss RE, Schleicher JB. Application of a multiple surface tissue culture propagator for the production of cell monolayers, virus, and biochemicals. Biotech and Bioengr 1968; 10: 617-24.

19. Urokinase-Pulmonary Embolism Trial Study Group, Urokinase pulmonary embolism trial, phase I results. JAMA 1970; 214: 2163-72.

20. Urokinase-Pulmonary Embolism Trial Study Group, Urokinase-streptokinase embolism trial, phase II results. JAMA 1974; 229: 1606-13.

21. American Heart Association. 2001 Heart and Stroke Statistical Update. Dallas: American Heart Association, 2000.

22. Leng GC, Papacosta O, Whincup P, *et al.* Femoral atherosclerosis in an older British population: prevalence and risk factors. Atherosclerosis 2000; 152: 167-74.

23. Criqui MH, Fronek A, Barett-Connor E. The prevalenceof peripheral arterial disease in a defined population. Circ 1985; 71: 510-5.

24. Newman AB. Peripheral arterial disease: insights from population studies of older adults. J Geriatr Soc 2000; 48: 1157-62.

25. Donaldson MC, Mannick JA, Whittemore AD. Femoro-distal bypass with in situ greater saphenous vein: long-term results using the Mills valvulotome. Ann Surg 1991; 213: 457-65.

26. Bosch J, Hunink M. Meta-analysis of the results of percutaneous transluminal angioplasty and stent placement for aortoiliac occlusive disease. Radiology 1997; 204: 87-96.

27. Blaisdell F, Steele M, Allen R. Management of acute lower extremity arterial ischaemia due to embolism and thrombosis. Surg 1978; 84: 822-31.

28. Jivegard L, Holm J, Schersten K. The outcome in arterial thrombosis misdiagnosed as arterial embolism. Acta Chir Scand 1986; 12: 251-6.

29. Yeager RA, Moneta GL, Taylor LM, Jr., *et al.* Surgical management of severe acute lower extremity ischemia. J Vasc Surg 1992; 15: 385-93.

30. Mills JL, Porter JM. Acute limb ischemia. In Porter JM, Taylor LM, Jr., eds. Basic data underlying clinical decision making in vascular surgery. St. Louis: Quality Medical Publishing, Inc., 1994. pp. 134-6.

31. Englund R, Magee HR. Periheral arterial embolism: 1961-1985. Aust N Z J Surg 1987; 57: 27-31.

32. Fogarty TJ, Daily PO, Shumway NE, *et al.* Experience with balloon chatheter technic for arterial embolectomy. Am J Surg 1971; 122: 231-7.

33. Cambria RP, Abbott WM. Acute arterial thrombosis of the lower extremity: its natural history contrasted with arterial embolism. Arch Surg 1984; 119: 784-7.

34. McNamara TO. Thrombolysis as the initial treatment for acute lower limb ischemia. In Comerota AJ, ed. Thrombolytic Therapy for Peripheral Vascular Disease. Philadelphia: J.B. Lippincott Company, 1995. pp. 253-68.

35. Whittemore AD, Donaldson MC, Mannick JA. Ten-year patency of autogenous vein bypass grafts. In Yao JST, Pearce W, eds. Long-term results in vascular surgery. Norwalk: Appleton & Lange, 1993. pp. 243-6.

36. Erickson CA, Towne JB, Seabrook GR, *et al.* Ongoing vascular laboratory surveillance is essential to maximize lont-term in situ saphenous vein bypass patency. J Vasc Surg 1996; 23: 18-27.

37. Ouriel K, Shortell CK, DeWeese, JA, *et al.* A comparison of thrombolytic therapy with operative revascularization in the initial treatment of acute peripheral arterial ischemia. J Vasc Surg 1994; 19: 1021-30.

38. Field T, Littooy FN, Baker WH. Immediate and long-term outcome of acute arterial occlusion of the extremities. Arch Surg 1982; 117: 1156-60.

39. Abbott WM, McCabe C, Maloney RD, *et al.* Embolism of the popliteal artery. Surg Gyne & Obstet 1984; 159: 553-6.

40. Faggioli GL, Ricotta JJ. Thrombolytic therapy for lower extremity arterial occlusion. Ann Vasc Surg 1993; 7: 297-302.

41. McNamara TO, Fischer JR. Thrombolysis of peripheral arterial and graft occlusions: improved results using high dose urokinase. AJR 1985; 144: 769-75.

42. STILE Investigators. Results of a prospective randomized trial evaluating surgery vs. thrombolysis for ischemia in the lower extremity. Ann Surg 1994; 220: 250-68.

43. Ouriel K, Veith FJ, Sasahara AA. A comparison of recombinant urokinase with vascular surgery as initial treatment for acute arterial occlusion of the legs. New Engl J Med 1998; 338: 1105-11.

44. Carter, TA. Clinical Internship with the Consumer Products Clinical Division at Alcon Laboratories, Inc.: Literature Review on Dry Eye Syndrome and associated Ocular Flora