CRYONIC SUSPENSION CASE REPORT: A-1165

Alcor #: A-1165
Patient Name:
Date of Birth: 9/9/1910
Date of Suspension: 07 October, 1988

Background History

Shortly after becoming a Suspension Member in October of 1986, Mr. ------ began talking with his mother about the desirability of her making cryonic suspension arrangements. There was some urgency to this request owing to Mrs. ------ poor health, her residence in an acute-care convalescent facility in Indianapolis, Indiana, and her long history of severe chronic obstructive pulmonary disease (COPD).

According to her son, Mrs. ------ initially expressed no interest in making suspension arrangements owing to her poor physical condition and long-standing depression. She reportedly expressed the sentiment that she "had no reason to live."

In August of 1988, her medical condition began to deteriorate and Mrs. ------ was again approached by her son regarding cryonic suspension arrangements. This time, according to her son, sensing that she was dying, she expressed interest in making suspension arrangements. Mrs. ------ is a Christian Scientist, but did not believe in an afterlife and, while her son states that she was not very sanguine about the prospects for cryonics being effective at saving at her life, she nevertheless felt it was better than no chance at all. Her son then began attempting to make the legal and financial arrangements necessary to provide for her cryonic suspension. In the process of doing this it was necessary for him to borrow money from an uncle who did not approve of cryonic suspension. This is mentioned here because a condition imposed by the uncle in providing the loan was that the uncle speak with Mrs. ------ and establish her consent. He did this, and the loan was made.

Subsequent to the completion of Mrs. ------ suspension arrangements, Steve Bridge, Alcor Midwest Coordinator, met with Mrs. ------ and discussed her interest in making cryonics arrangements and established her informed consent. Mr. Bridge described Mrs. ------ as lucid, and oriented as to person, place, and time when this conversation took place.

On 27 September, 1988 Mrs. ------ submitted a completed *Authorization For Anatomical Donation*, *Consent for Cryonic Suspension*, *Cryonic Suspension Agreement*, *Certificate of Religious Belief* and *Living Will*. Subsequently, on 27 September, 1988 Mrs. ------ submitted a completed *Application for Cryonic Suspension*, selecting the neurosuspension option. On 1 October, 1988 Mrs. ------ was approved as an Alcor Suspension Member.

Medical History

Mrs. ------ is a 78-year-old, widowed, caucasian female with oxygen and steroid dependent end-stage chronic obstructive pulmonary disease (COPD) (with a bronchospastic

component) secondary to tobacco abuse, chronic depression, wide angle glaucoma, prednisone-induced osteoporosis with multiple spontaneous vertebral fractures, and severe post-herpetic neuropathy. She is a resident of the --------- Retirement Home.

The patient had a 50 pack-year history of tobacco abuse and gave up smoking in 1979. In September of 1986, the patient was hospitalized with an episode of severe dyspnea which responded to therapy with bronchodialators and steroids. During this hospitalization she was completely worked up for COPD with the following findings: FVC and FEV_1 values (post bronchodialators administration) of 0.92 and 0.42 liters, respectively. Arterial blood gases on room air showed a pO_2 of 60 mmHg, a pCO_2 of 43, and a pH of 7.48. Radionuclide cardiac studies showed essentially normal left ventricular function and her EKG showed no specific changes. She was discharged in October of 1986 on 2 liters per minute (lpm) oxygen, theophylline, and Bronkosol.

Her pre-deanimation medical history is remarkable for multiple hospitalizations for respiratory acidosis, pneumonia, and air hunger.

Throughout 1988 the patient continued on a slow downhill course with increasing air hunger, unremitting pain from her post-herpetic neuralgia, and deterioration of her overall fitness which resulted in her confinement to bed.

Late in September of 1988 the patient's food intake began to diminish and by early October the patient was taking very little by mouth. The last few days of before deanimation the patient's p.o. intake consisted of a few ounces of water per day.

At 15:00 on 5 October 1988, the patient had a tarry black stool which tested Hematest positive indicating upper GI bleeding.

Pre-Transport Hematology and Chemistry

At the request of the Transport Team, the patient's physician, Dr. --- , ordered lab samples drawn at 14:30 on 6 October, 1988. The results were as follows:

Complete Blood Count

Test Procedure	*Results*
White Blood Cells (RBC)	17.4
Red Blood Cells (WBC)	3.67
Hematocrit (HCT)	36.9
Hemoglobin (Hgb)	11.6
Mean Corpuscular Volume (MCV)	100.5
Mean Corpuscular Hemoglobin (MCH)	31.6
Mean Corpuscular Hemoglobin Concentration (MCHC)	31.4
Lymphocytes	8%
Monocytes	2%
Segs	90%

RBC Morphology: size: normal, color: normal, shape: normal.
Platelet Count: 759,000 cu. mm.

Chemistries

mOsm	320.3 mOsm
SGOT	35 IU/l
SGPT	44 IU/l
Total Bilirubin	0.7 mg/dl
BUN	77.0 mg/dl
Creatinine	2.2 mg/dl
Cholesterol	186 mg/dl
Alkaline Phosphatase	236 IU/l
Glucose	107 mg/dl
Phosphorus	6.1 mg/dl
Calcium	8.3 mg/dl
Total Protein	5.5 g/dl
Albumin	3.4 g/dl
Globulin	2.1 g/dl
Sodium	150 mEq/l
Potassium	4.2 mEq/l
Chloride	104 mEq/l
CO_2	22 mEq/l
Uric Acid	10.5 mg/dl
Lactate Dehydrogenase	248 IU/l

Deanimation

On 4 October at 19:30 the patient's condition began to deteriorate markedly, with increasing air hunger and rising pulse. Blood presure declined from 108/40 to 60/40 by 08:00 the following day, with pulse remaining fairly steady at about 120. Vital signs starting from 10:00 through to deanimation are presented in tabular and graphic form below:

Time	*Blood Pressure*	*MAP*	*Pulse (Carotid)*	*Respirations*	*Observations*
10/5/88					
10:00	60/30	40	120	16	resting quietly
14:45	50/p	---	120	16	resp. shallow
22:00	120/60	80	116	24	resp. labored
10/6/88					
07:00	60/p	---	120-5	N/A	resp. labored
08:00	50/0	---	80	20	oriented x 3
21:30	110/60	76	102	36	resting quietly

10/7/88

01:30	62/40	47	80	36	color poor
06:00	40/p	---	76	36	expirations "grunty"
12:00	0/0	---	80	24	no pulses in extremities, mucous membranes cyanotic, agonal respirations.
01:20	0/0	---	0	0	deanimation

At approximately 11:00 on 7 October, following the IM administration of 50 mg demerol and 25 mg phenergan for pain, the patient's respirations began diminishing in frequency and she became tachycardic and diaphoretic. Her blood pressure became unobtainable by cuff either by auscultation or palpation. At 12:00 it was noted that peripheral pulses had disappeared, the patient was cyanotic, and agonal respirations were present. At 13:20 the patient experienced cardiac arrest in the --------- Retirement Home in Indianapolis, Indiana.

Transport

A Remote Standby Transport Team from Alcor Southern California (ASC) consisting of Mike Darwin and Jerry Leaf, and a local standby team consisting of Steve Bridge and Robert Schwartz was standing by.

An Esophageal Gastric Tube Airway (EGTA) was placed by the Alcor Transport Team upon pronouncement of cardiac arrest by the attending Registered Nurse (R.N.) in the nursing home and manual cardiopulmonary support (CPS) was begun using a bag-valve respirator employing 100% oxygen, 5:1 compression to ventilation ratio, at a rate of 60 compressions per minute. CPS was briefly interrupted upon arrival of the physician at approximately 14:05 (ca. 30 second interruption) so that legal death could be pronounced. Legal death was pronounced at 14:08.

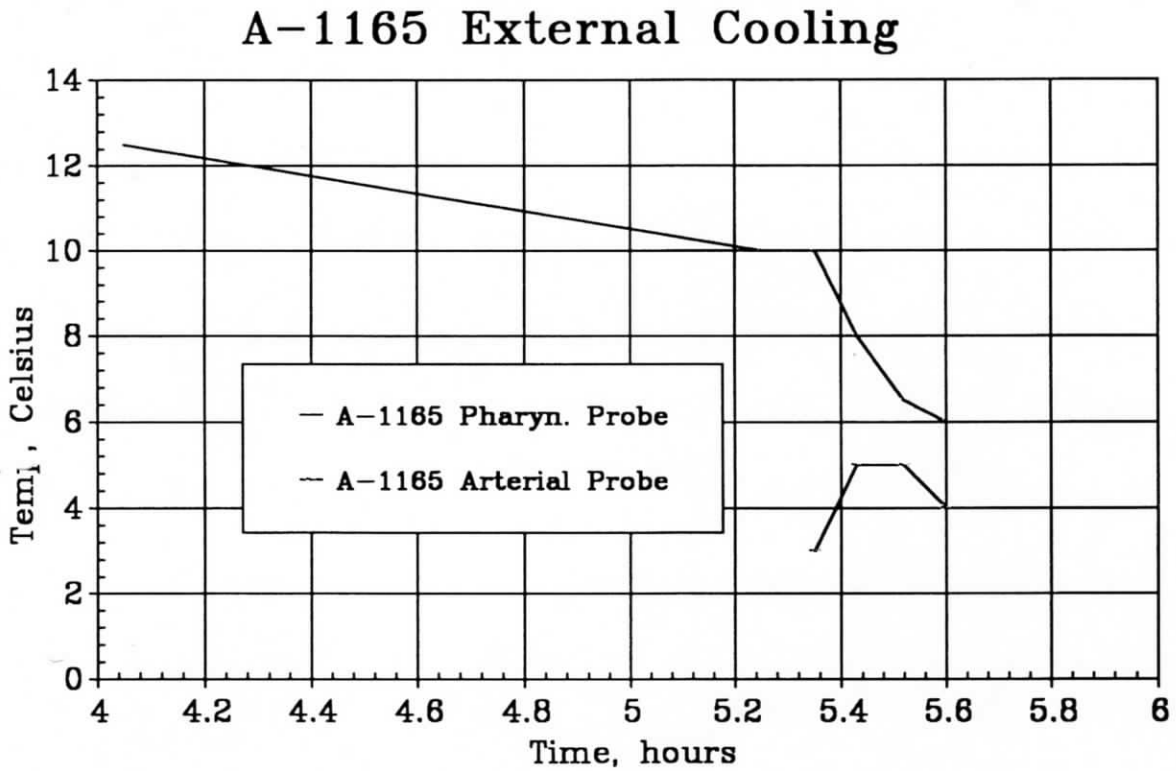
Mechanical cardiopulmonary support (CPS) was begun at 14:12 using a Brunswick Heart-Lung Resuscitator (HLR) 50-90: 60 compressions/min. 1-1/2" sternal deflection (85 lb. force setting) with twelve 1500 cc ventilations per minute via EGTA. The patient was then transferred to an ambulance cot, removed from the hospital, and loaded into a mortuary van for transport to the Royster-Askin-Sandrock mortuary where total body washout was to be carried out.

External cooling was begun at 13:40 using Zip-Loc polyethylene bags containing crushed ice. Administration of transport medications began at 14:21. All transport medications were given IV as follows: 39.4 mEq potassium chloride at 14:21, 1 gm deferoxamine at 14:20, 16,584 I.U. sodium heparin at 14:19; 11.8 mg verapamil at 14:20, 78.8 gm mannitol in 500 cc water at 14:23; 2.76 mg metubine iodide at 14:23; 36 gm tromethamine (THAM). Infusion at 14:19.

Mechanical cardiopulmonary support and infusion of THAM was continued en route to the mortuary. THAM administration was completed shortly after arrival at approximately 15:30.

At approximately 15:00, frankly bloody gastric contents were noted to be issuing from the gastric tube of the EGTA. One hundred twenty-five cc of bloody stomach contents were suctioned.

Due to malfunction of two of the telethermometers in the Remote Standby Kit, temperature monitoring was delayed until the patient's arrival at the mortuary. The first temperature obtained was a pharyngeal temperature of 12.5°C at 17:28. Temperature was monitored with a Yellow Springs Instrument Co. (YSI) Model 43TA thermistor thermometer employing YSI type 401 vinyl coated thermistor probes.



A venous blood sample drawn at 17:16 from the peripheral I.V. catheter used to give the transport medications (first 5 cc discarded) disclosed the following:

Complete Blood Count

Test Procedure	*Results*
White Blood Cells (RBC)	3.8
Red Blood Cells (WBC)	1.90
Hematocrit (HCT)	16.9
Mean Corpuscular Volume (MCV)	89
Mean Corpuscular Hemoglobin (MCH)	29.3
Mean Corpuscular Hemoglobin Concentration (MCHC)	33.1
Bands	00.0 per 100 WBC
Neutrophils	63%
Lymphocytes	36%
Monocytes	1%
Eosinophils	0%
Basophils	0%

RBC Morphology: 2+ macrocytosis, 1+ polychromasia, no spherocytes observed.
Platelet Estimate: platelets appear slightly decreased.

Chemistries

mOsm	446
pH	7.17
SGOT	1040 IU/l
SGPT	726 IU/l
Total Bilirubin	0.3 mg/dl
Direct Bilirubin	0.1 mg/dl
Indirect Bilirubin	0.2 mg/dl
BUN	98.0 mg/dl
Creatinine	3.3 mg/dl
Cholesterol	92 mg/dl
Alkaline Phosphatase	139 IU/l
Glucose	19 mg/dl
Phosphorus	11.7 mg/dl
Calcium	6.6 mg/dl
Total Protein	3.3 g/dl
Albumin	1.4 g/dl
Globulin	1.9 g/dl
Sodium	113 mEq/l
Potassium	29.2 mEq/l
Chloride	90 mEq/l
CO_2	12 mEq/l
Creatine Phosphokinase	268 IU/l
gamma-GT	181 IU/l
Uric Acid	22.0 mg/dl
Lactate Dehydrogenase	2172 IU/l
Amylase	<25 IU/l
Lipase	169 IU/l

Total Body Washout Perfusate Preparation

The composition of the total body washout flush solution is given in Table I. Dry chemical perfusate components were prepared from reagent or medical grade chemicals weighed out using an Ohaus Centogram model 311, or Ohaus Triple Beam 2610 g balances. Dry components were mixed with sterile water for injection USP, or sterile water for irrigation USP. Perfusates were sterilized by filtration into the oxygenator of the extracorporeal circuit through a Pall PP3802 0.20μ pre-bypass filter. A total of 20 liters of washout perfusate was prepared.

TABLE I

SHP-1 Total Body Washout Perfusate

Component	*Molar Concentration (mM)*	*g/l*
Sucrose	170 (MW 342.30)	58.19
Glucose	10.0	1.80
HEPFS (Na^+ salt)	7.2	3.90
Glutathione	5.0	0.92
Sodium Bicarbonate	10.0	0.84
Potassium Chloride	23.7	1.34
Calcium Chloride	1.0	0.11
Magnesium Chloride	2.0	0.095
Hydroxvethyl Starch	---	50.0
Heparin	---	1000 units/l

pH was to 8.02.

mOsm: 295 (measured).

NOTE: Due to the omission of potassium chloride (KCl) from the dry components it was necessary to use liquid potassium chloride injection which was present in the Remote Standby Kit in preparing the perfusate. Since an inadequate amount of liquid KCl was available to make up the deficit, 1 liter of Plasmalyte was substituted for 1 liter of water for injection to raise the osmolality to an acceptable level. Plasmalyte has the following composition per liter: 5.26 gm sodium chloride, 5.02 gm sodium gluconate, 3.68 gm anhydrous sodium acetate, 370 mg potassium chloride, and 300 mg magnesium chloride.

Total Body Washout (TBW)

Femoral Cannulation

The patient's right groin was prepared for femoral cut-down by scrubbing/swabbing with povidone iodine solution (Betadine) and draping with sterile towels. The anatomical position of the right femoral artery and vein were located by reference to the pubic tubercle and the anterior superior iliac spine. An incision with a #10 scalpel blade was made at the midpoint between these two structures, beginning with the inguinal ligament and running parallel to the longitudinal axis of the leg for approximately 5 cm.

The femoral artery and vein were dissected free and #2 silk ties placed on proximal and distal exposures of both vessels. The distal ties were tied to achieve occlusion. There was a visible arterial pulse from the HLR.

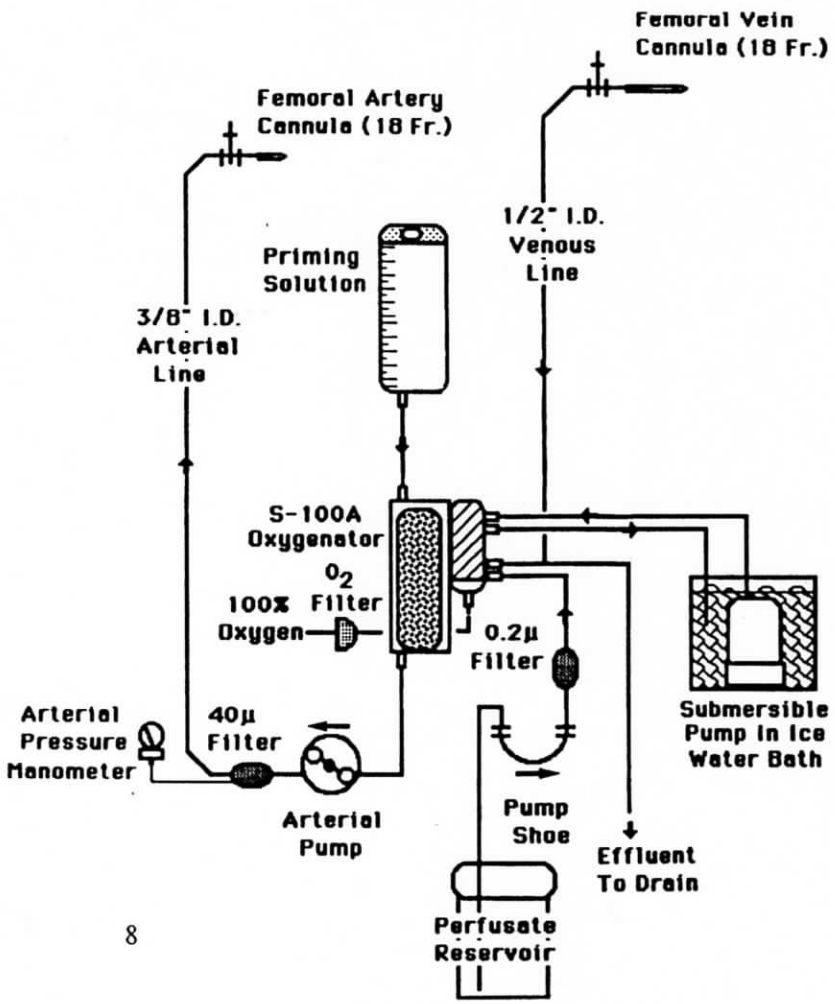
An arteriotomy was made with with a #11 scalpel blade and a USCI Type 1966, 18 Fr. arterial cannula was introduced and secured with the proximal tie. A veinotomy was performed in the same fashion and a USCI type 1967, 30 Fr. venous cannula was advanced until the tip was well within the inferior vena cava near the heart and secured with the proximal tie. Blood from the arteriotomy was bright red and appeared to be well oxygenated.

The arterial perfusion line was connected to the arterial cannula with a 3/8" straight connector with port, and the port fitted with a Cobe 3-way stopcock for evacuation of air.

The venous return line was connected to the venous cannula with a 1/2" straight connector with port, and air removed from the venous cannula and venous line with a 35 cc plastic syringe.

Femoral-femoral partial bypass was initiated at 18:43 in the preparation room of the mortuary employing a custom-built, 2-head roller pump, Shiley S-100A bubble oxygenator, and a Shiley SAF-20, 20μ blood filter. Perfusion pressure was measured at the Shiley SF-20 filter, anterior to the arterial cannula, employing an aneroid manometer with a sterile Tri-Med **Isolator** flexible membrane barrier to protect the aneroid from fluid contamination. A calibration curve of measured back-pressure versus measured flow was generated in advance to account for the pressure increase resulting from cannula-induced flow restriction. The extracorporeal circuit is presented schematically below:

TBW Extracorporeal Circuit



The extracorporeal circuit was primed with 1 liter of Plasmalyte, 500 cc of Dextran 40, 44.6 mEq of sodium bicarbonate (50 cc), and 2500 I.U. of sodium heparin. The composition of Plasmalyte is given in Table II. Bypass was initiated at a blood flow rate of 2.25 liters per minute and an oxygen flow rate of 4.0 liters per minute. Trans-cannula arterial pressure at the start of bypass was 140 mmHg (measured before the cannula). Adjusting for pressure drop across the cannula yields a systemic perfusion pressure of 56 mmHg. At the start of extracorporeal circulation, ventilation and chest compression were discontinued and the mask of the Esophageal Gastric Tube Airway (EGTA) was removed (the obturator was left in place to guard against aspiration). Closed circuit extracorporeal circulation was continued for approximately nine minutes.

TABLE II

Plasmalyte

Component	*g/l*	
Sodium Chloride	5.26	water for injection qs.
Sodium Acetate	3.68	
Sodium Gluconate	5.02	294 mOsm/l (calculated)
Potassium Chloride	0.37	
Magnesium Chloride	0.30	pH adjusted to 5.5 with hydrochloric acid

Blood Washout

Washout with SHP-1 perfusate (composition given in Table I) began at 18:52 with the patient's temperature at approximately 8°C and consisted of eight liters of base perfusate. A second pass of 12 liters was begun 18:55 at a pressure of 64 mmHg and concluded at 19:03. The arterial pump flow rate averaged three liters per minute. A venous effluent sample drawn from the venous line at 18:52 near the beginning of the first exchange disclosed the following:

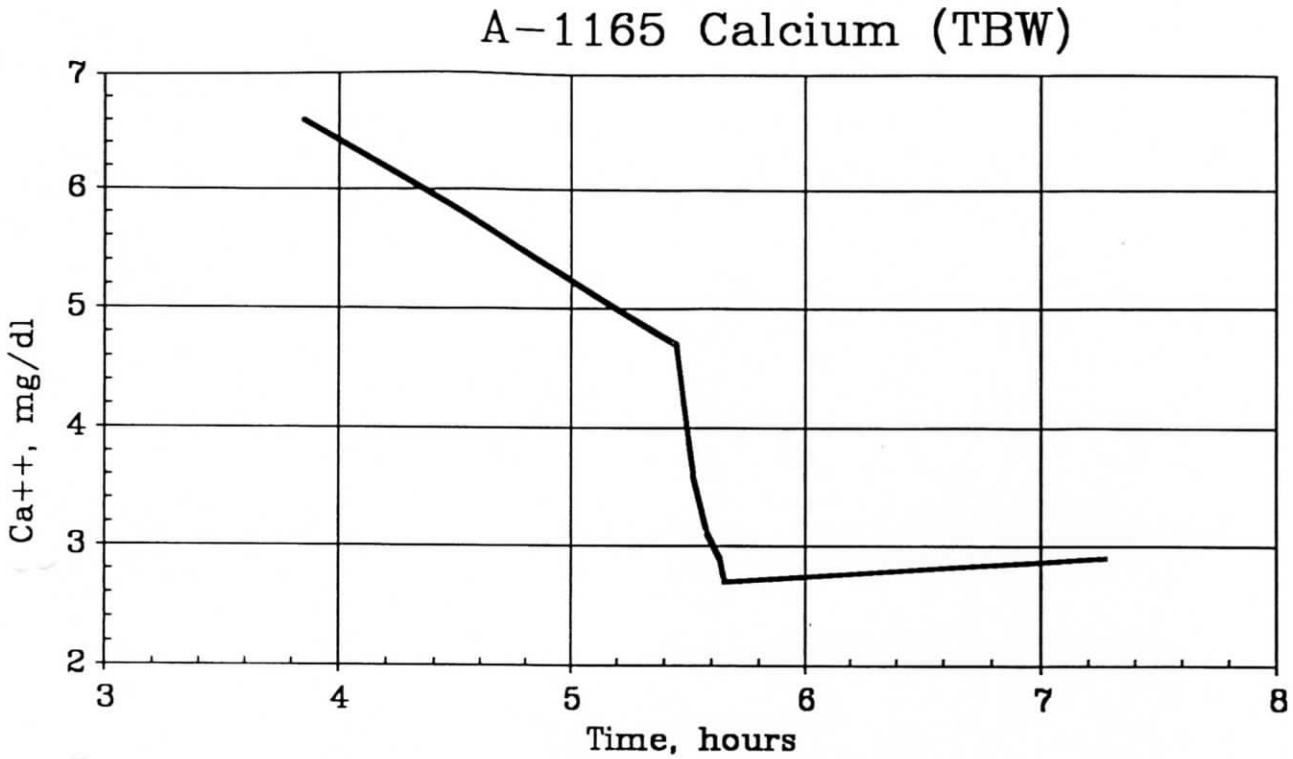
pH	6.94
mOsm	389
HCT	---
SGOT	725 IU/l
SGPT	461 IU/l
Total Bilirubin	0.2 mg/dl
Direct Bilirubin	0.1 mg/dl
Indirect Bilirubin	0.1 mg/dl
BUN	70.0 mg/dl
Creatinine	2.4 mg/dl
Cholesterol	35 mg/dl
Alkaline Phosphatase	117 IU/l
Glucose	40 mg/dl
Phosphorus	10.7 mg/dl
Calcium	4.7 mg/dl
Total Protein	1.7 g/dl
Albumin	0.6 g/dl
Globulin	1.1 g/dl
Sodium	132 mEq/l

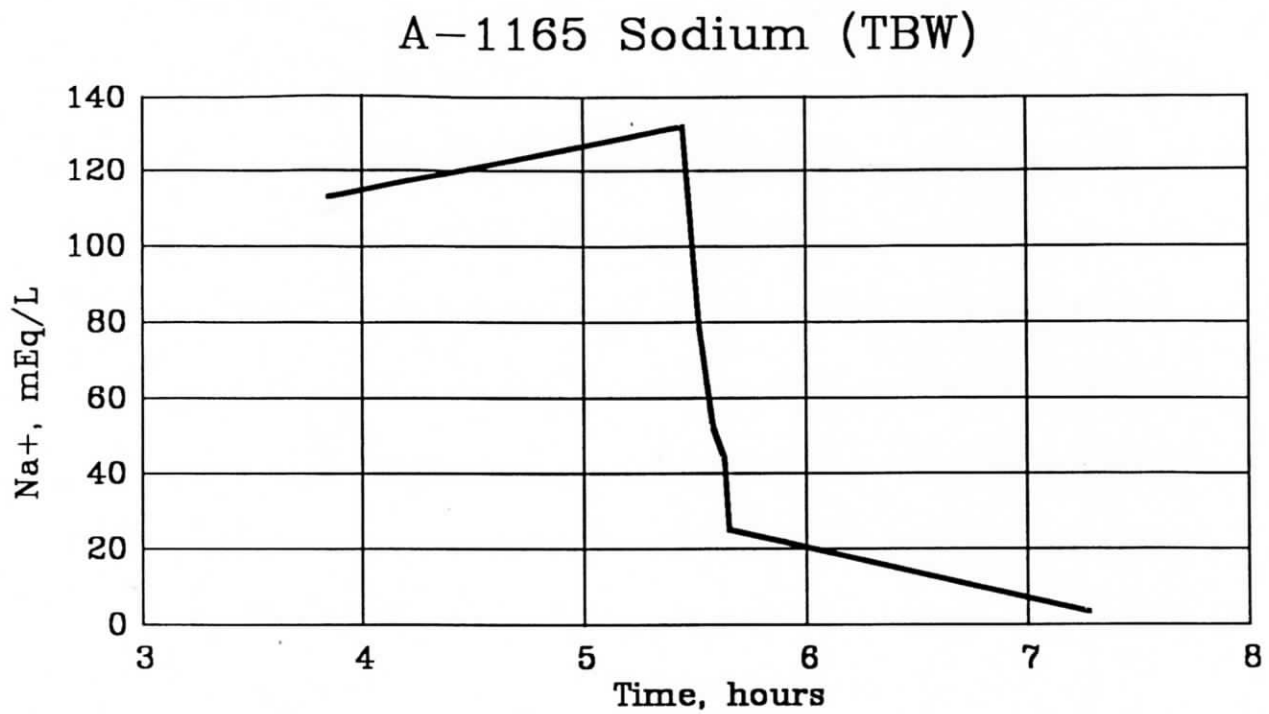
Potassium	19.2 mEq/l
Chloride	98 mEq/l
CO_2	9 mEq/l
Creatine Phosphokinase	94 IU/l
gamma-GT	83 IU/l
Uric Acid	12.7 mg/dl
Lactate Dehydrogenase	1510 IU/l
Amylase	<25 IU/l
Lipase	56 IU/l

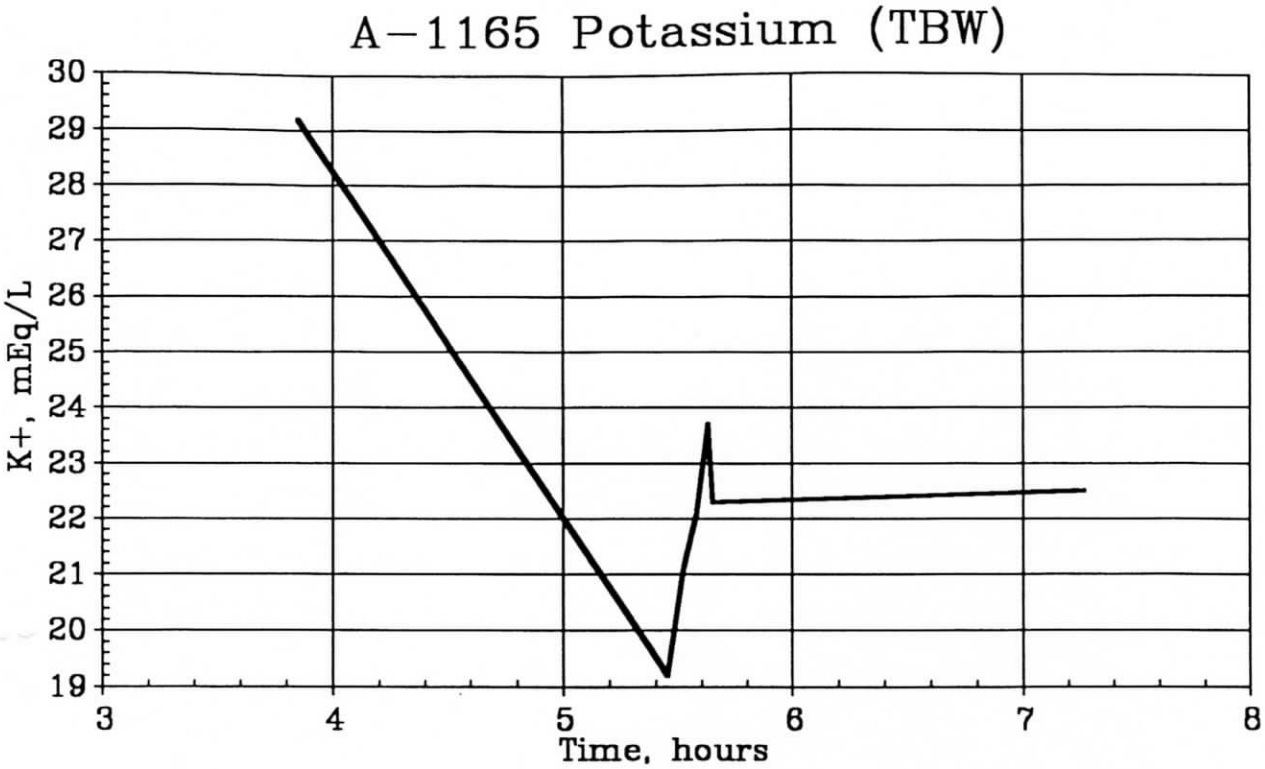
A venous effluent sample taken at 19:00, near the conclusion of TBW, disclosed the following:

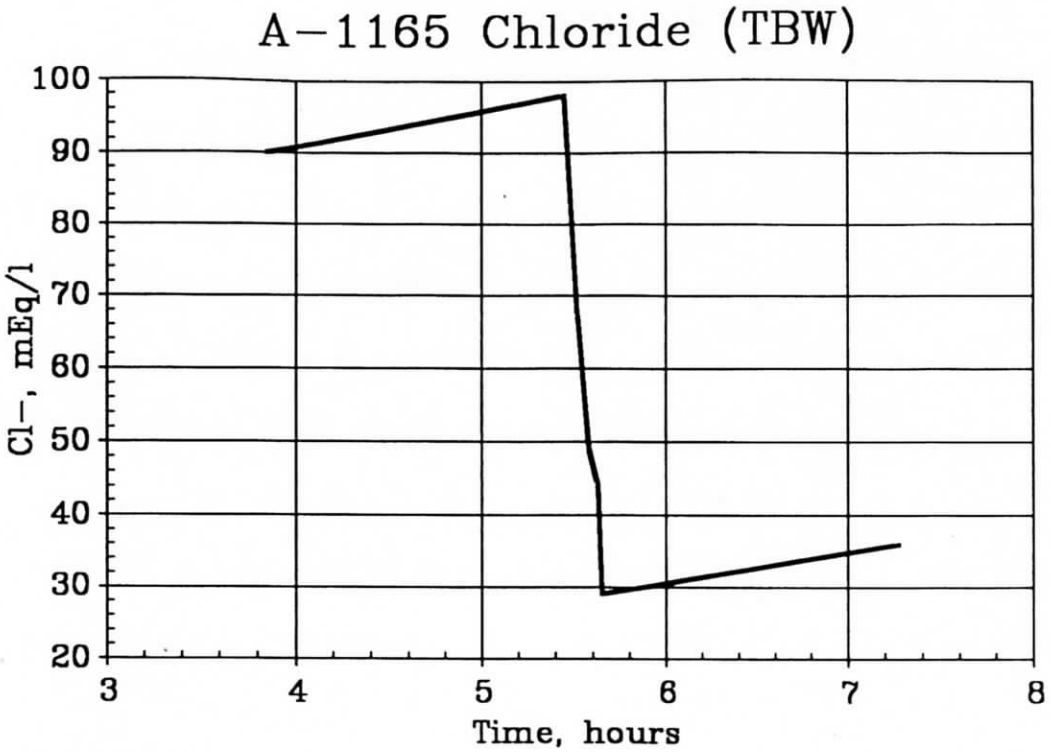
pH	7.08
mOsm	346
SGOT	106 IU/l
SGPT	68 IU/l
Total Bilirubin	0.1 mg/dl
Direct Bilirubin	0.0 mg/dl
Indirect Bilirubin	0.1 mg/dl
BUN	27.0 mg/dl
Creatinine	0.9 mg/dl
Cholesterol	0 mg/dl
Alkaline Phosphatase	7 IU/l
Glucose	259 mg/dl
Phosphorus	2.5 mg/dl
Calcium	3.1 mg/dl
Total Protein	1.7 g/dl
Albumin	0.0 g/dl
Globulin	1.7 g/dl
Sodium	52 mEq/l
Potassium	22.1 mEq/l
Chloride	49 mEq/l
CO_2	6 mEq/l
Creatine Phosphokinase	6 IU/l
gamma-GT	4 IU/l
Uric Acid	0.8 mg/dl
lactate Dehydrogenase	282 IU/l
Amylase	<25 IU/l
Lipase	28 IU/l

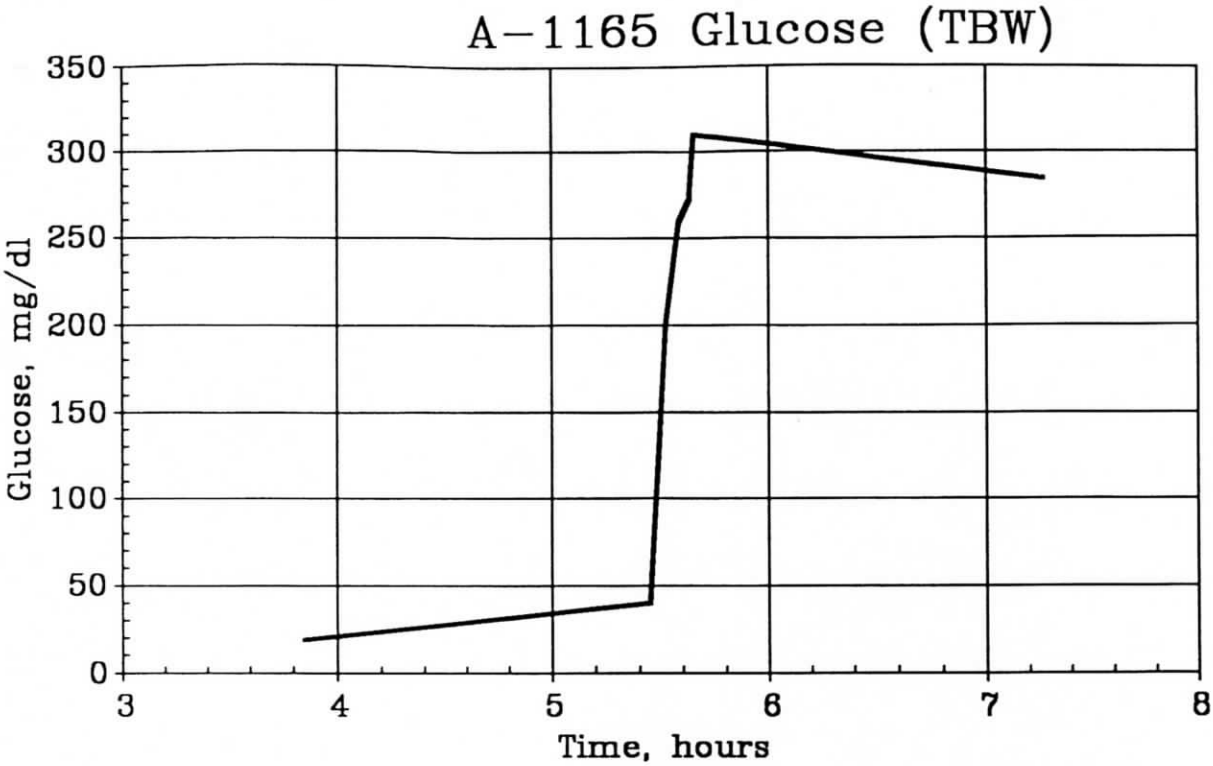
Laboratory evaluations of samples taken during TBW are presented graphically below:

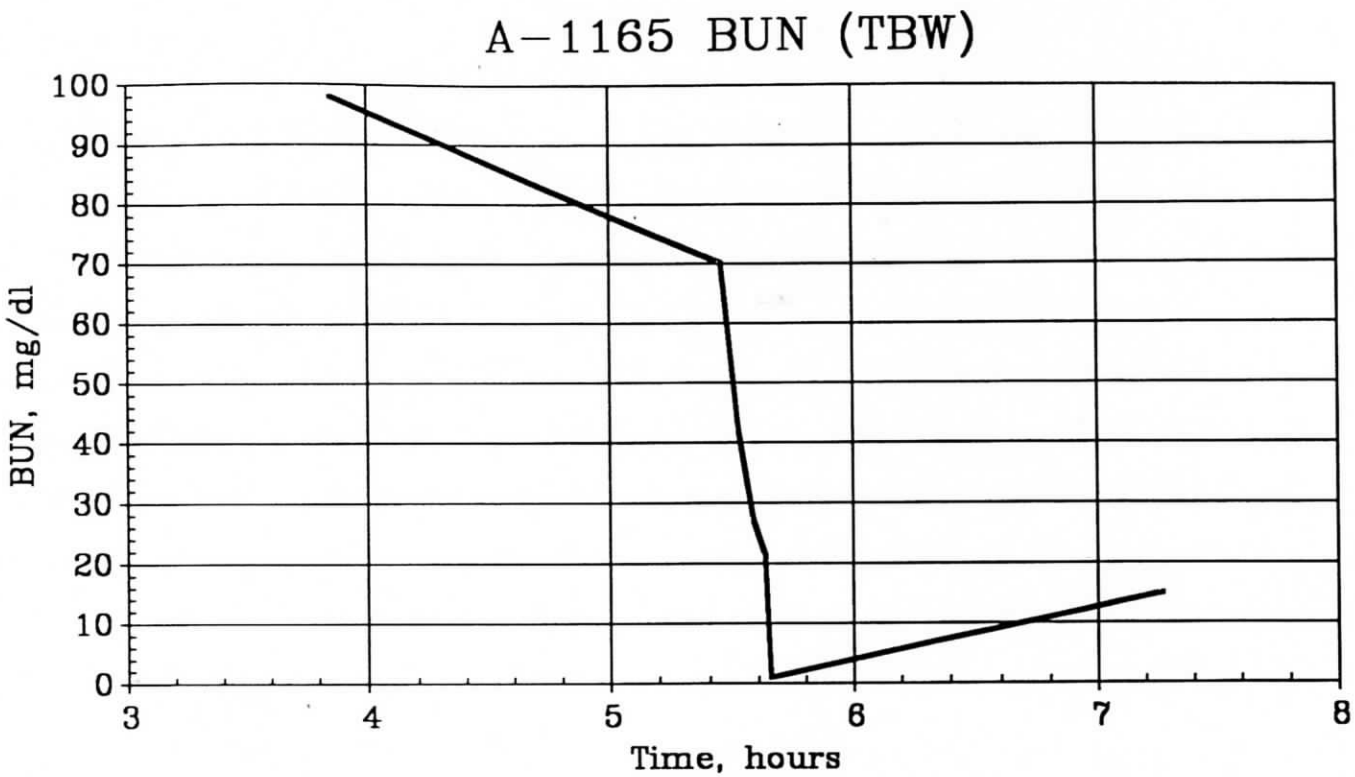




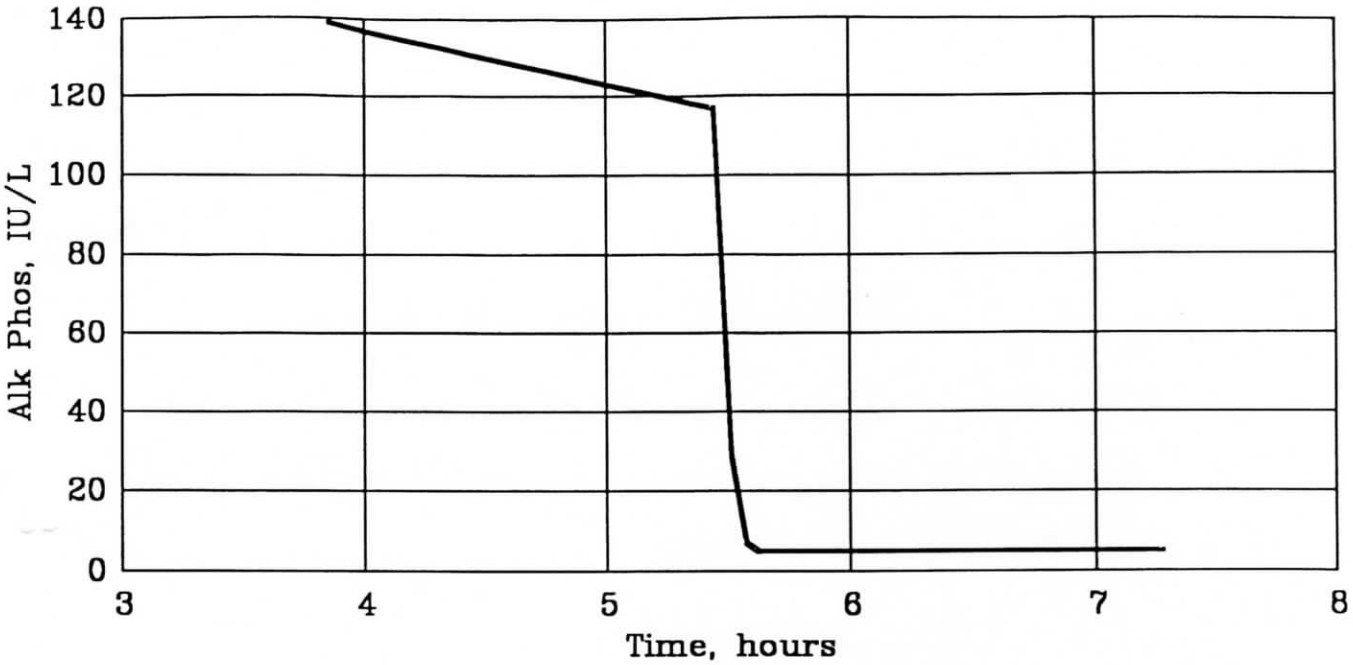

A−1165 Potassium (TBW)
K+, mEq/L
30
29
28
27
26
25
24
23
22
21
20
19
3
4
5
6
7
8
Time, hours


A−1165 Chloride (TBW)
Cl−, mEq/l
100
90
80
70
60
50
40
30
20
3
4
5
6
7
8
Time, hours

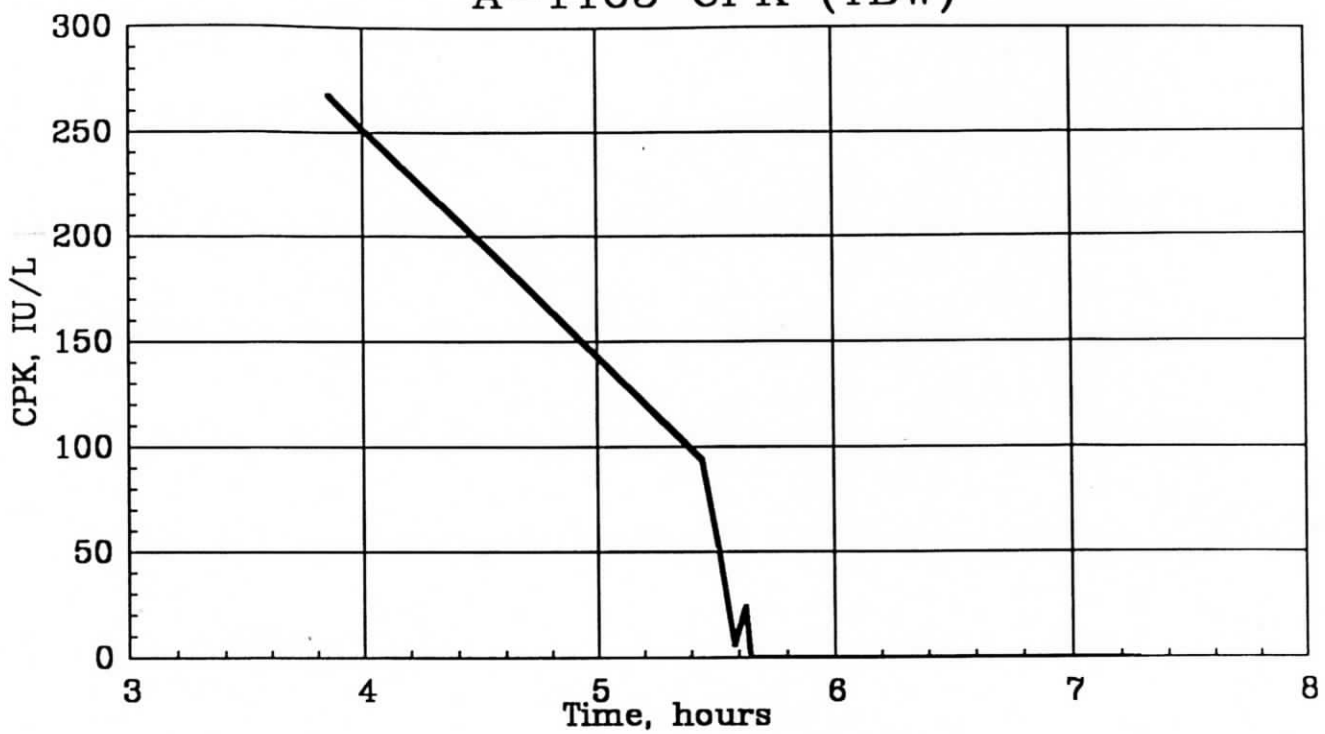

A−1165 Glucose (TBW)
Glucose, mg/dl
350
300
250
200
150
100
50
0
3
4
5
6
7
8
Time, hours

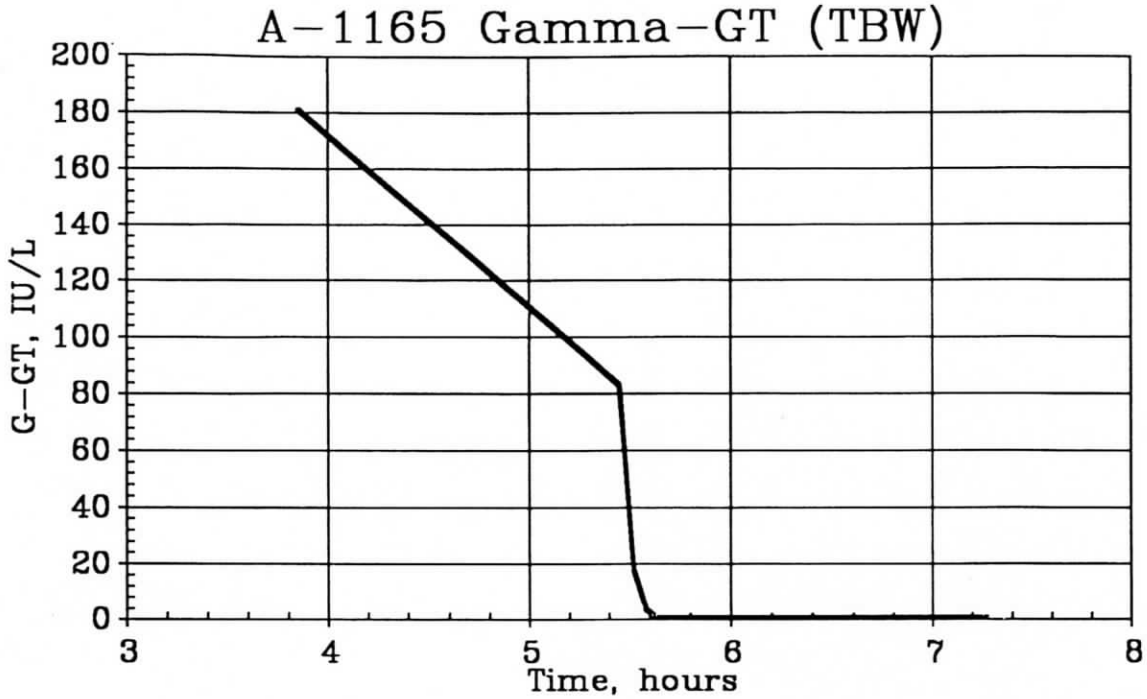

A−1165 BUN (TBW)
BUN, mg/dl
100
90
80
70
60
50
40
30
20
10
0
3
4
5
6
7
8
Time, hours

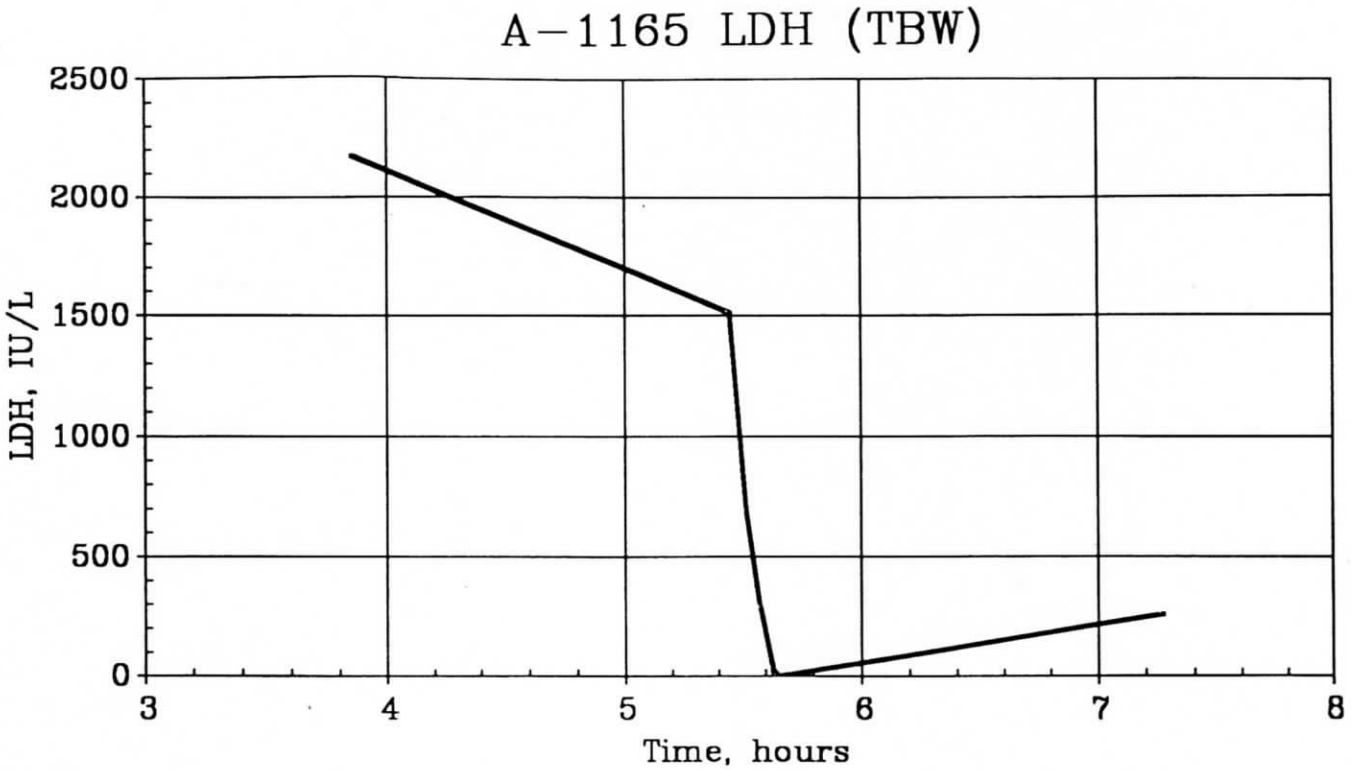
A-1165 Alkaline Phosphatase (TBW)

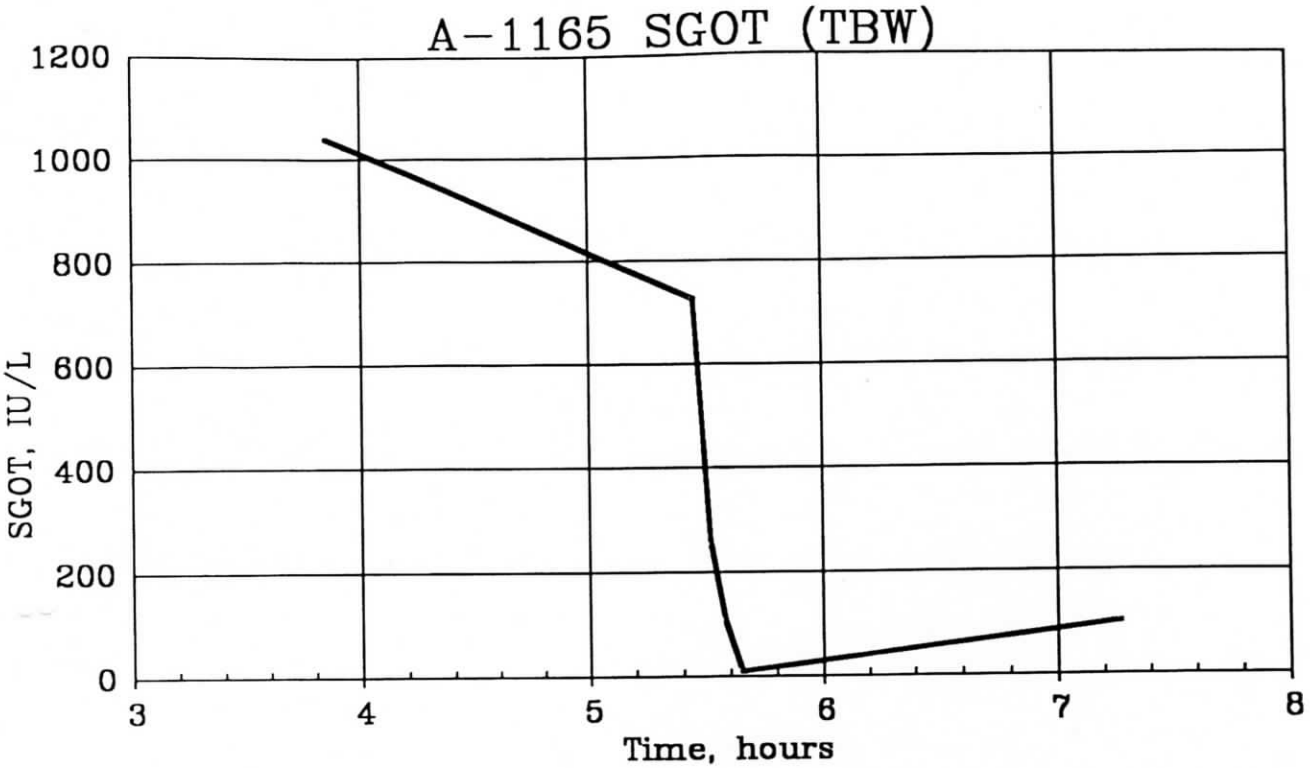


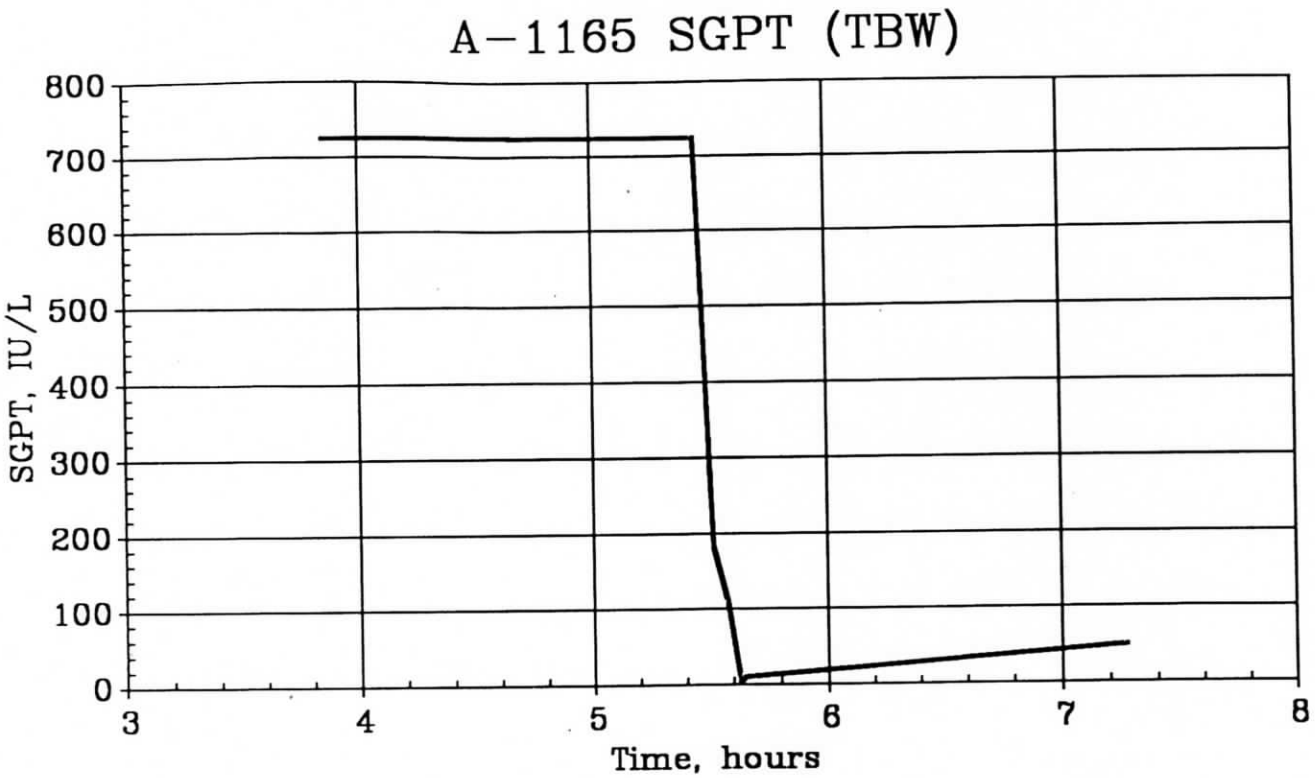
A-1165 CPK (TBW)




A−1165 Gamma−GT (TBW)
G−GT, IU/L
200
180
160
140
120
100
80
60
40
20
0
3
4
5
6
7
8
Time, hours


A−1165 LDH (TBW)
LDH, IU/L
2500
2000
1500
1000
500
0
3
4
5
6
7
8
Time, hours


A-1165 SGOT (TBW)
1200
1000
800
600
400
200
0
SGOT, IU/L
3
4
5
6
7
8
Time, hours


A-1165 SGPT (TBW)
800
700
600
500
400
300
200
100
0
SGPT, IU/L
3
4
5
6
7
8
Time, hours

TBW was concluded at 19:03 at an esophageal temperature of 5.8°C and the patient was quickly cleaned up on the prep table and transferred to a heavy-duty vinyl body bag. At this time (19:17) it was noted that the jaws were in rigor. A quick assessment of the patient disclosed no other muscles in rigor at that time. The body bag containing the patient was then placed atop a bed of Zip-Loc bags containing crushed ice which had been laid down inside an insulated air transport box. The patient was then covered with additional bags of crushed water ice and the transport container was closed for air shipment to Alcor's perfusion facility in Riverside, California.

The next commercial flight to Southern California from Indianapolis was not available until the following morning. The patient, packed in ice, remained at the mortuary overnight.

Air Transport

Air shipment was on U.S. Air Flight #93, which arrived at Ontario Airport at approximately 12:50 on 8 October, 1988. Air transport was uneventful. The patient was collected from the freight handling department of U.S. Air and transported to the Alcor facility by Cryovita van, arriving at 13:40.

Gross Assessment

The patient's arrival temperatures were: 1.6°C esophageal and 1.2°C rectal. The patient was transferred from the air shipment container to the operating table, the surface of which had been previously prepared with a cooling blanket placed atop 2"-thick eggcrate foam. After the patient was on the operating table she was briefly examined. The exam disclosed a thin, caucasian female in her late 70's. The arms and calves were thin, the abdomen was grossly normal. The eye exam disclosed bilaterally dilated pupils with corneal misting. The buccal mucosa was gray-white and apparently blood-free. The skin was pale, uniform in color, and apparently blood-free. There were scattered petechiae on the upper extremities and occasional petechiae on the trunk and lower limbs. The skin under the HLR piston was bruised and erythematous in appearance. The distal half of the sternum exhibited the "caved in" appearance which is typical following extended HLR support.

The muscles of the jaw, left arm, and lower extremities were noted to be in rigor. The neck, right forearm, and right hand were observed to be free of rigor.

Perfusate Preparation

The composition of the perfusate employed to carry out cryoprotective perfusion is given in Table III. Dry chemical perfusate components were prepared from reagent or medical grade chemicals weighed out using an Ohaus Centogram model 311, and Ohaus Triple Beam 2610 g balances. Dry components were mixed with American Chemical Society (ACS) reagent grade glycerol and/or sterile water for injection USP, or sterile water for irrigation USP. Perfusates were sterilized by filtration into the recirculating and cryoprotective concentrate reservoirs through a Pall PP3802 0.20μ pre-bypass filter. Perfusate was prepared in two batches with the following volumes and glycerol concentrations:

Description	Volume	Glycerol%(w/v)
Recirculating	29 liters	5%
Concentrate	20 liters	72%

TABLE III

SHP-1 Cryoprotective Perfusate

Component	*Molar Concentration (mM)*	*g/l*
Sucrose	170 (MW 342.30)	58.19
Glucose	10.0 (MW 180.2)	1.80
HEPES (Na^+ salt)	7.2 (MW 260.3)	3.90
Glutathione	5.0 (MW 307.3)	0.92
Sodium Bicarbonate	10.0 (MW 84.0)	0.84
Adenine HCl	1.0 (MW 180.6)	0.17
Ribose	.94 (MW 150.2)	0.14
Potassium Chloride	28.3 (MW 74.56)	2.11
Calcium Chloride	1.0 (MW 111)	0.11
Magnesium Chloride	2.0 (MW 95.2)	0.095
Dextran 40	--- (MW 45,000)	50.0
Heparin		1000 units/l

pH was 7.90 (measured) mOsm: 299 (not measured).

Operative Procedures

Pre-operative Prep

The patient was prepared for a median sternotomy and cranial burr-hole by shaving the head and thorax and scrubbing/swabbing them with povidone iodine solution (Betadine). The sternal operative site was defined by draping with sterile towels and an adhesive operative drape (3M) was placed over the sternum. A cardiac drape was placed over the patient, "tented" on two IV poles at the head and allowed to extend down over the feet and over the sides of the table by a minimum of 24". The top of the scalp was draped with three surgical drapes to define a triangular operative site over the right frontal lobe.

Median Sternotomy/Vascular Access

Median sternotomy commenced at 18:26 with an incision over the midline of the sternum with a #10 scalpel blade. Fascia and connective tissue were cleared down to the sternum with an electrosurgical knife. A median sternotomy was then performed with Mayo scissors. The edges of the sternotomy were padded with laparotomy sponges, a self-retaining retractor placed, and the sternotomy retracted open. Blunt and sharp dissection were used to expose the pericardium. The ascending aorta was freed from the pulmonary artery by blunt dissection with Metzenbaum scissors. An aortic cross-clamp was placed just above

the aortic valve to exclude the coronary circulation. A second aortic cross-clamp was applied to the descending aorta just distal to the left subclavian artery in order to exclude any arterial circulation to the body. Tourniquets (cable ties) were applied to each arm just below the axilla to sequester the arms from flow.

A ventral midline pericardiotomy was made using Metzenbaum scissors. Four stay sutures of 3-0 silk were placed in the margins of the pericardiotomy. These sutures were tied to the sternal retractor, thereby reflecting the pericardium away, and creating a pericardial "cradle" and exposing the heart and aorta for cannulation. A Sarns cardiotomy sucker was used to suction away the pericardial fluid.

A 3-0 Tycron purse-string suture was placed in the aorta and a snare applied. An aortotomy was made with a #11 scalpel blade. A 22 Fr. aortic perfusion cannula was primed with normal saline and a clamp placed on the distal end. The cannula was then introduced into the aorta and snared in place with a hemostat.

A Satinsky partial occlusion clamp was placed on the right atrium just below the apex. A purse-string suture of 2-0 Tycron was placed in the atrium and a snare tube applied. An atriotomy was made by removing the apex of the right atrium with Metzenbaum scissors. A tube clamp was placed on the distal end of the 32 Fr. USCI type 1967 venous catheter and it was advanced through the atriotomy (with concurrent release of the Satinsky clamp) into the right atrium to the superior vena cava. Umbilical tape was passed around the superior vena cava and tied below the cannula tip. In order to prevent contamination of the recirculating system with venous circulation from the extremities, silk ties were placed on the left and right innominate veins just distal to the left and right internal jugular veins. Venous return was collected from the cannula in the superior vena cava.

A third small purse-string suture of 5-0 silk was was placed in the left lateral aspect of the ascending aorta and an aortotomy made with a #11 scalpel blade. A Cobe 3-way stopcock was fitted to an Aloe arterial pressure monitoring catheter, and the catheter was flushed with normal saline and introduced through the aortotomy into the ascending aorta. The catheter was secured in place by applying a snare to the 5-0 suture.

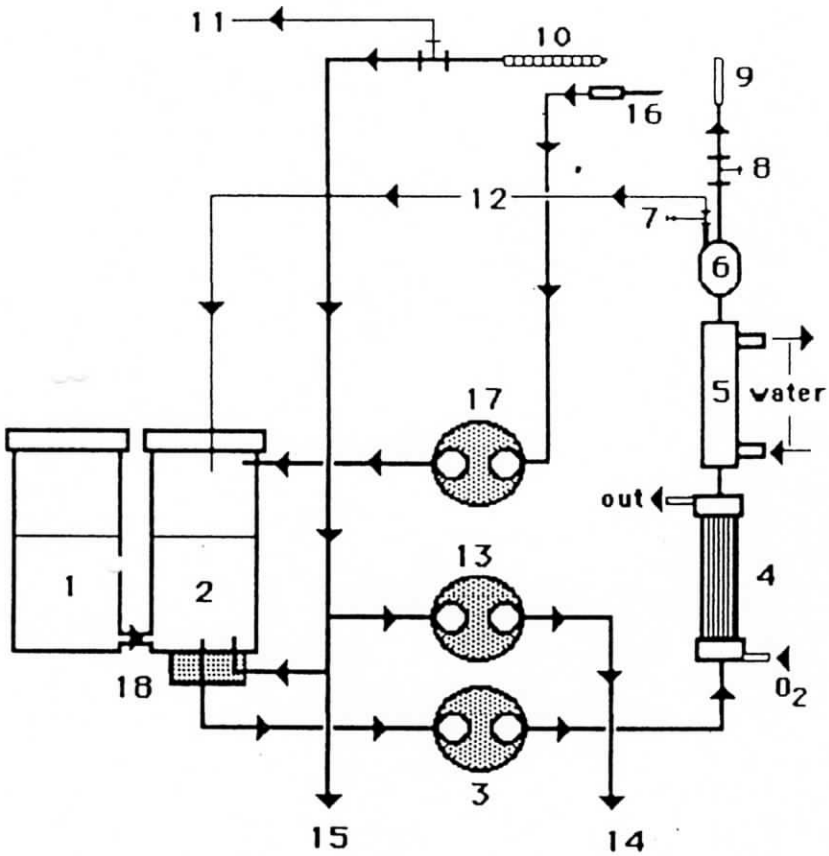
The sterile perfusion tubing was then brought up to the surgical field and secured in a Travenol tubing holder towel clamped to the drapes. The arterial-venous loop of the perfusion circuit was clamped and divided by cutting out the 1/2" - 3/8" adapter with Mayo scissors. A 1/2" connector with a Cobe 3-way stopcock was used to connect the 1/2" ID venous return line to the venous cannula. Air was cleared from the system with a 100 cc glass syringe. A Cobe 8 ft. pressure monitoring line was fitted to the arterial pressure catheter, flushed with normal saline and handed off the field to be connected to the Trantec Model 800 pressure transducer and Statham SC1001 monitor.

Surgery to connect the patient to the perfusion circuit was completed at approximately 20:00.

Perfusion Circuit

The extracorporeal circuit for cryoprotective perfusion is shown schematically below. The circuit consisted of two parts: a recirculating system to which the patient was connected, and a cryoprotectant addition system which was connected to the recirculating system. The recirculating system was a 20 liter reservoir sitting atop a magnetic stirring table, an arterial (recirculating) roller pump, an Erika HPF 300 hemodialyzer which was used as a hollow fiber oxygenator, a Sarns Torpedo heat exchanger, and a Pall

EC1440 40 micron blood filter. The recirculating (mixing) reservoir was continuously stirred with a 2" teflon-coated magnetic stirring bar driven by a Thermolyne type 7200 magnetic stirrer. The cryoprotectant addition system consisted of a 20-liter polyethylene reservoir containing 50% (w/v) glycerol (see Table I) and a Drake-Willock model #7401 hemodialysis pump acting as a withdrawal pump which removed perfusate from the recirculating system, causing 50% (w/v) glycerol perfusate from the concentrate reservoir to flow under gravity into the recirculating reservoir.



CRYOPROTECTIVE PERFUSION CIRCUIT

1) Cryoprotective Concentrate Reservoir
2) Recirculating Reservoir
3) Arterial Pump
4) Oxygenator (HPF Dialyzer)
5) Heat Exchanger (Sarns)
6) 40μ Filter (Pall)
7) Sample Port (Arterial)
8) Connector with port
9) Cannula (Arterial)
10) Cannula (Venous)
11) Sample Port (Venous)
12) Vent Line (Arterial)
13) Withdrawal (Ramp) Pump
14) Withdrawal Discard Line
15) Total Body Washout Discard Line
16) Cardiotomy Sucker
17) Cardiotomy Suction Pump
18) Magnetic Stirring Table

Arterial and venous samples for evaluation of chemistries and glycerol concentration were drawn at 15-minute intervals during cryoprotective perfusion. Arterial samples were drawn from a 3-way stopcock interposed between the arterial filter and the filter vent line. Venous samples were drawn from a 8' Cobe monitoring line connected to a Cobe 3-way stopcock attached to the venous connector connecting the venous cannula and the venous return line. (The dead-space of the Cobe monitoring line was determined and this volume was drawn up and discarded before each sample was taken.)

The perfusion circuit was prepared in advance of need and was sterilized with ethylene oxide using an appropriate protocol of post-sterilization outgassing and aeration.

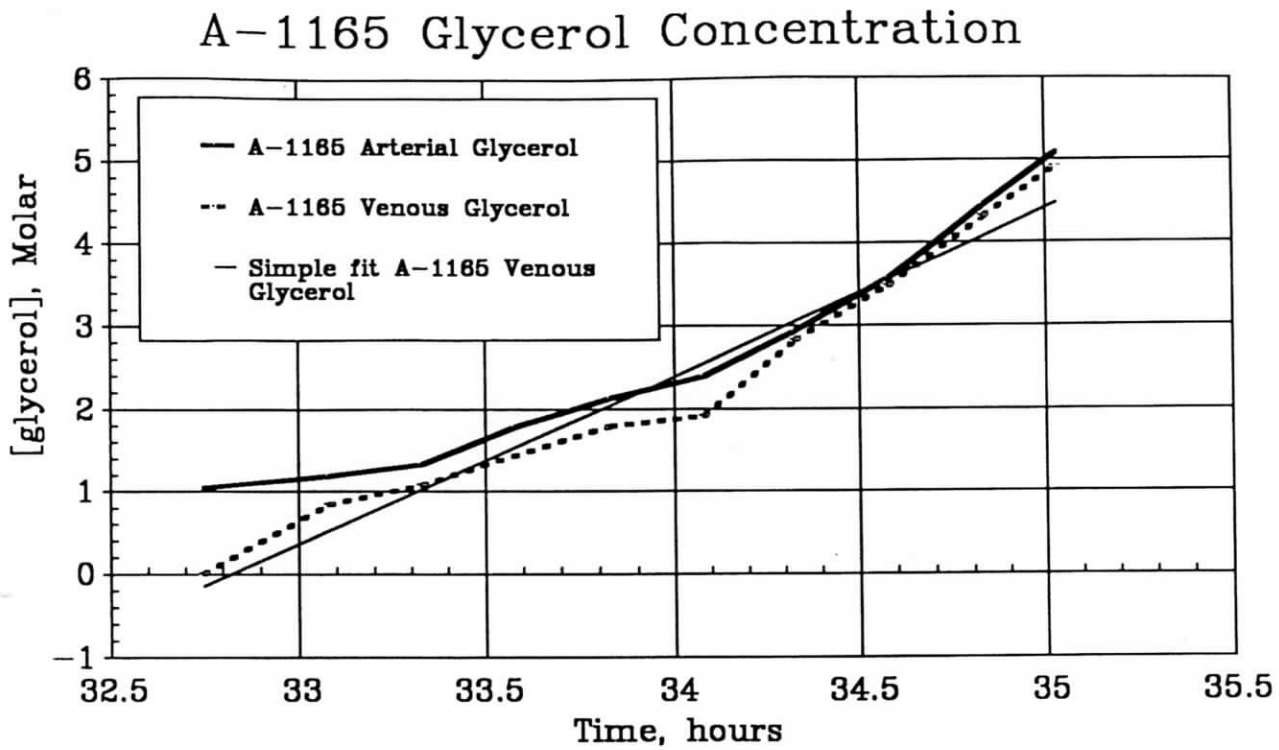
Cryoprotective Perfusion

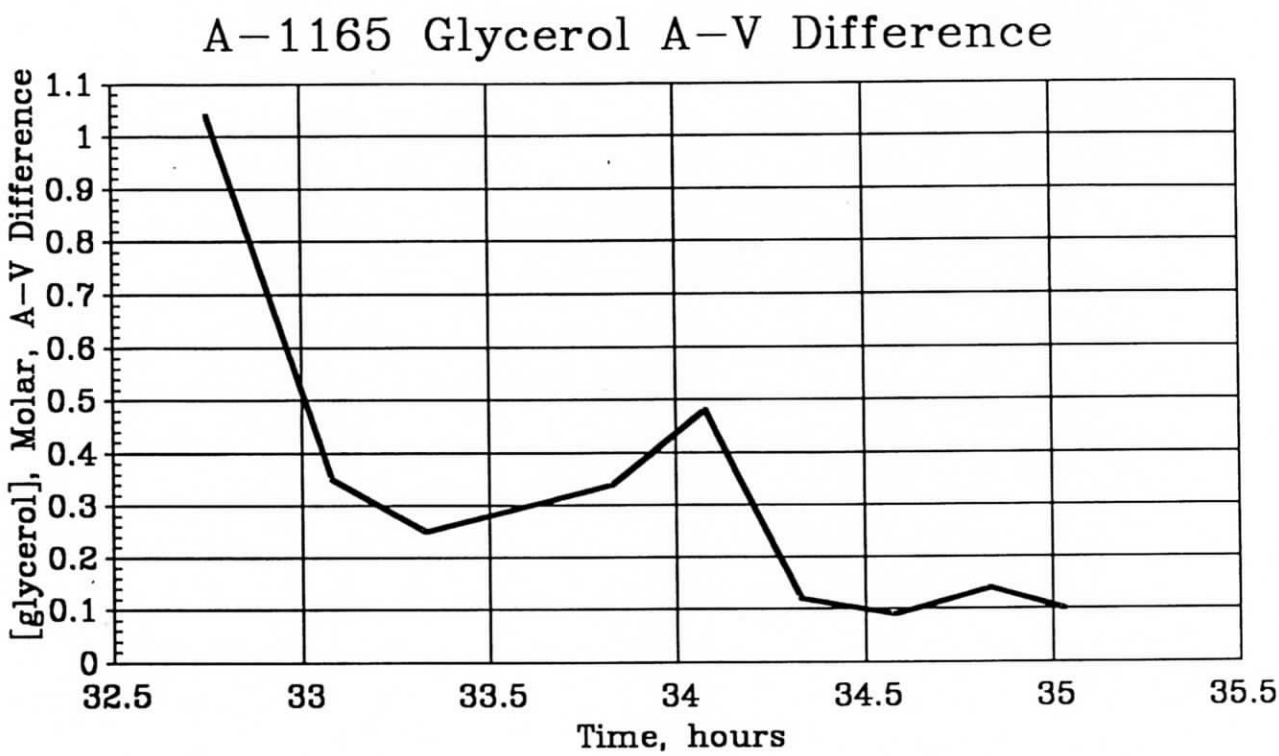
Because of the prolonged period of cold ischemia and the likely poor quality of perfusion during cardiopulmonary support, a decision was made to use nitrogen rather than oxygen during cryoprotective perfusion. Nitrogen flow to the oxygenator was 2.0 liters throughout perfusion. Closed-circuit perfusion of base perfusate was begun at 20:04 at a flow rate of 500 cc/min., a pharyngeal temperature of 3.5°C, an arterial temperature (perfusate) of 7.2°C and a mean arterial pressure (MAP) of 50 mmHg. Venous pH was measured at 7.17 at 20:10. At 20:21 glycerolization of the face and scalp was noted to be very uniform with no patchy areas of non-perfusion noted. Circulation through the scalp was judged good when it was incised for the burr hole; modest drainage (8-10 cc/min.) of clear perfusate was noted from the burr-hole throughout perfusion, presumably as a result of leakage from the scalp wound, craniotomy, and incised dura. At 21:30 the perfusion pressure (MAP) had increased to 55 mmHg and the arterial flow arat was reduced to 400 cc/min. to maintain arterial pressure near 50 mmHg. At 22:00 the MAP had risen back to 60 mm Hg and the arterial flow rate was again decreased, this time to 350 cc/min. A few minutes later, at 22:03 the arterial flow rate was further decreased to 300 cc/min. in an effort to hold the MAP under 60 mm Hg. Finally, at 22:10 the arterial flowrate was reduced to 250 cc/min. where it remained for the rest of the perfusion.

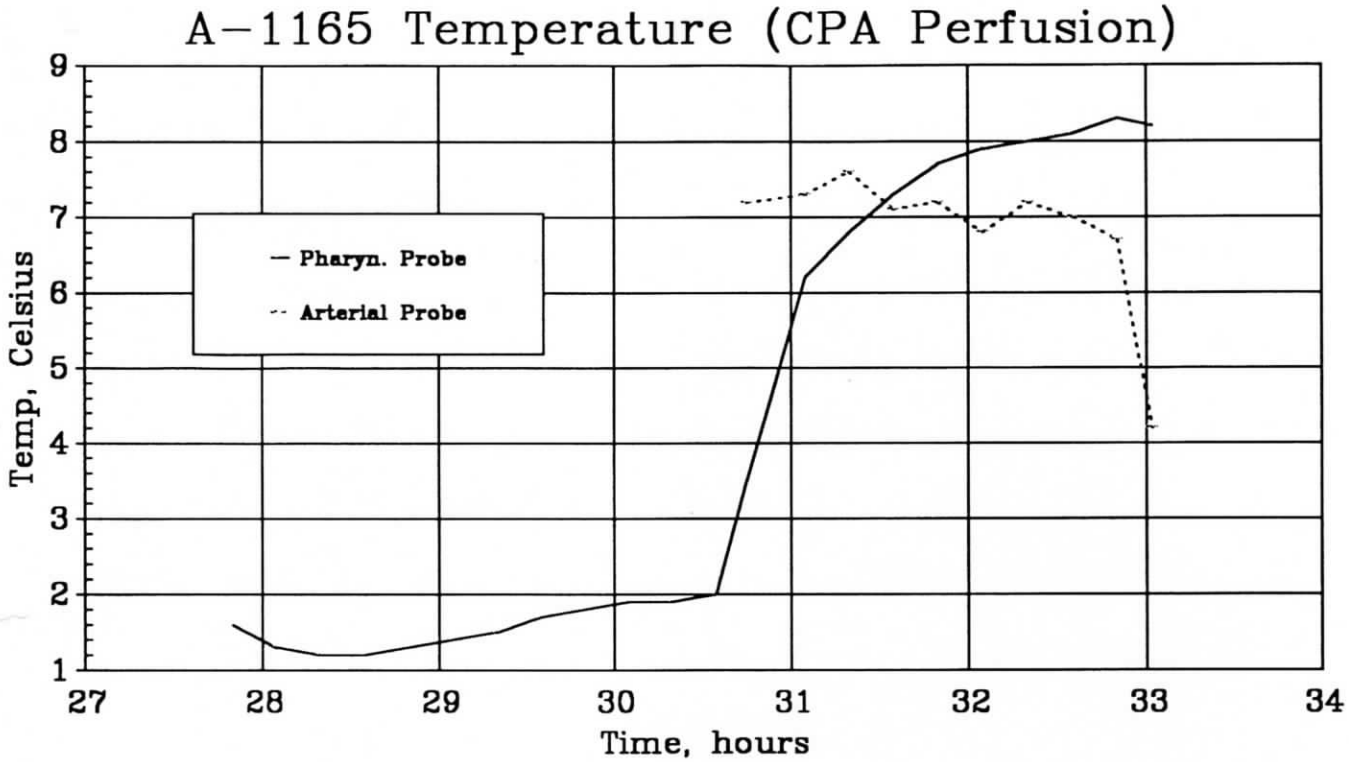
Cryoprotective perfusion was concluded at 22:41 at a flow rate of 250 cc/min., MAP of 61 mmHg, pharyngeal temperature of 8.2°C, sinus temperature of 7.4°C and an arterial temperature of 4.2°C.

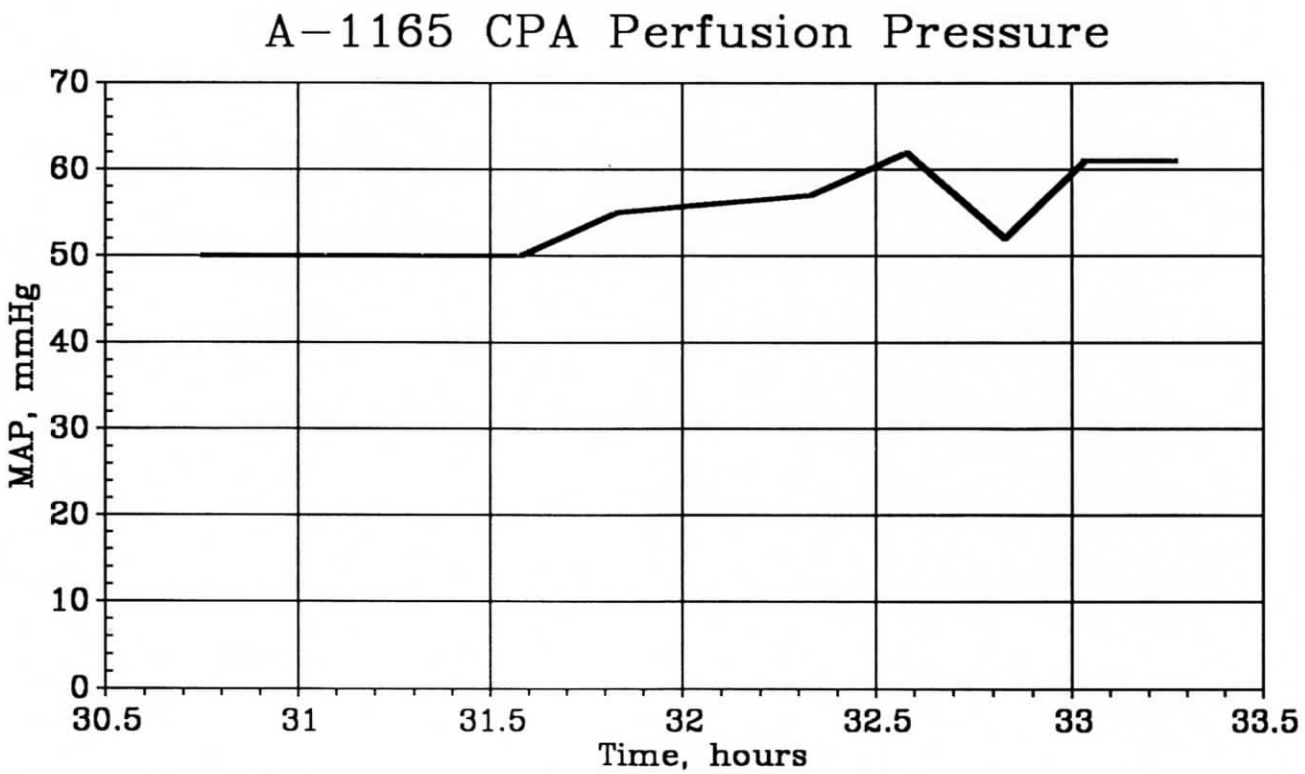
Cryoprotective Ramp

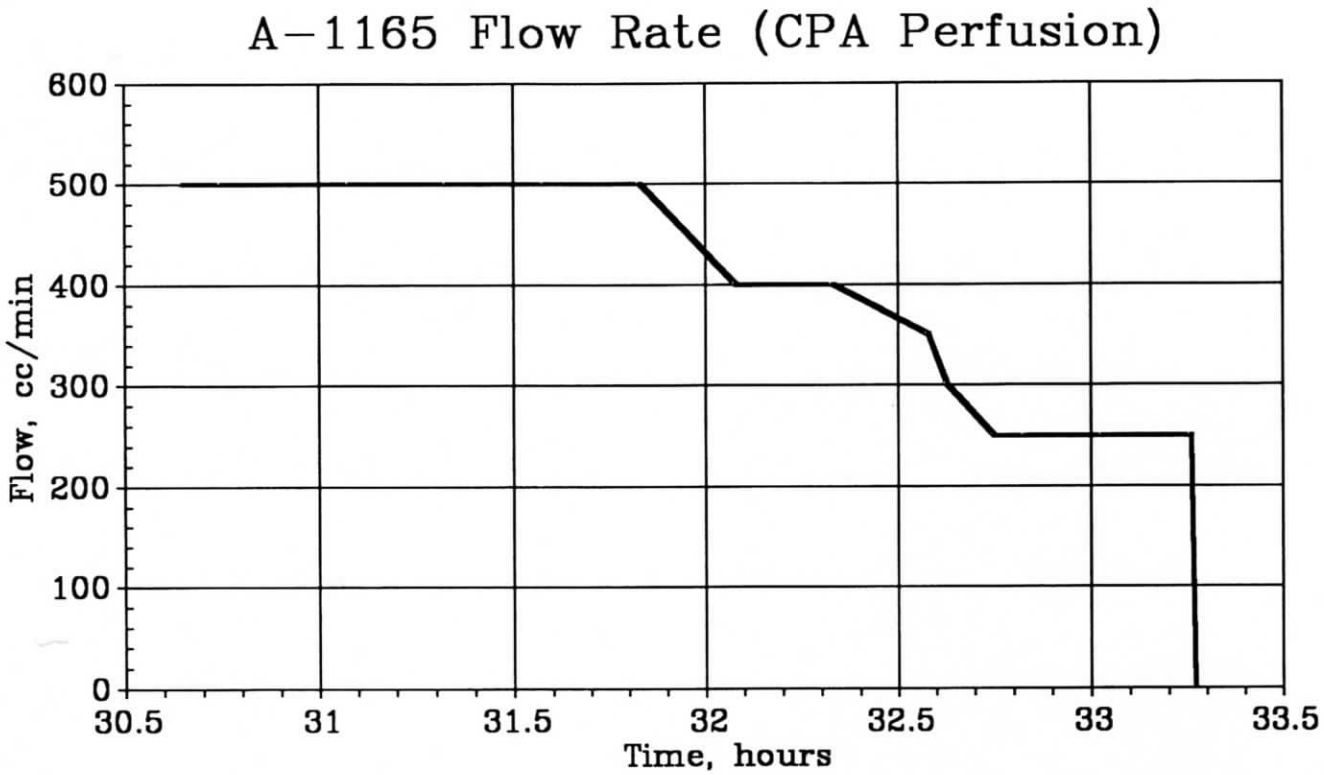
The recirculating perfusate withdrawal/glycerol concentrate addition flow rate was initially set at 100 cc/min. to obtain a rate of increase of cryoprotectant concentration of approximately 30 mM/min. At 21:30 the perfusate withdrawal/glycerol concentrate addition flowrate was increased to 150 cc/min., and at 21:45 to 200 cc/min. in response to developing cerebral edema. Despite these reactive changes in both the recirculation rate and the cryoprotectant addition rate, the rate of increase of cryoprotectant remained reasonably constant over the course of the perfusion, averaging 34 mM/min. Terminal glycerol concentration was 4.91 M in the final venous sample and 5.03 M in the final arterial sample. Glycerol concentration was determined refractometrically with an American Optical model 10400 Goldberg hand-held refractometer. The concentration of glycerol in the arterial and venous lines, a-v glycerol difference, arterial and esophageal temperatures, arterial pressure, and arterial flow rate during cryoprotective perfusion are shown graphically on the next page. Times shown are decimal hours post-arrest.

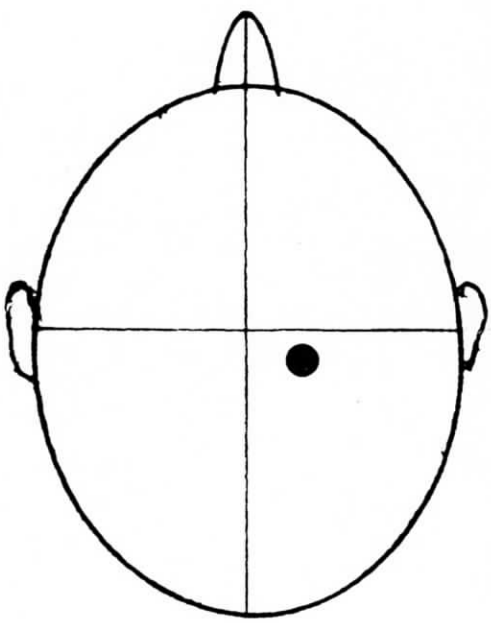

A−1165 Glycerol Concentration
A−1165 Arterial Glycerol
A−1165 Venous Glycerol
Simple fit A−1165 Venous Glycerol
[glycerol], Molar
Time, hours
6
5
4
3
2
1
0
−1
32.5
33
33.5
34
34.5
35
35.5


A−1165 Glycerol A−V Difference
[glycerol], Molar, A−V Difference
Time, hours
1.1
1
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0
32.5
33
33.5
34
34.5
35
35.5


A−1165 Temperature (CPA Perfusion)
Temp, Celsius
Time, hours
— Pharyn. Probe
-- Arterial Probe
1
2
3
4
5
6
7
8
9
27
28
29
30
31
32
33
34


A−1165 CPA Perfusion Pressure
MAP, mmHg
Time, hours
0
10
20
30
40
50
60
70
30.5
31
31.5
32
32.5
33
33.5





Cranial Burr-Hole

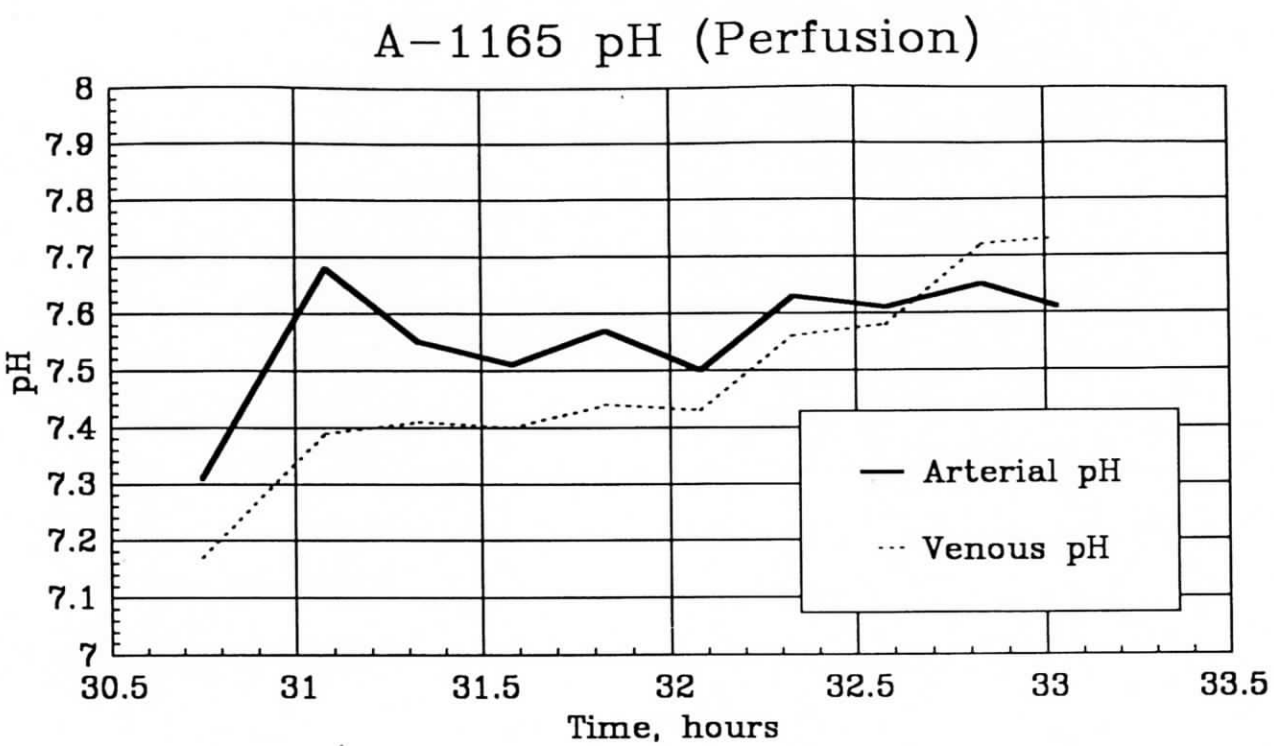
Surgery to open the cranial burr-hole was begun at 20:30. The vertex of the scalp approximately 3 cm to the right of midline over the right frontal lobe was incised with a #10 scalpel blade and an incision approximately 4 cm long was made down to the periosteum.

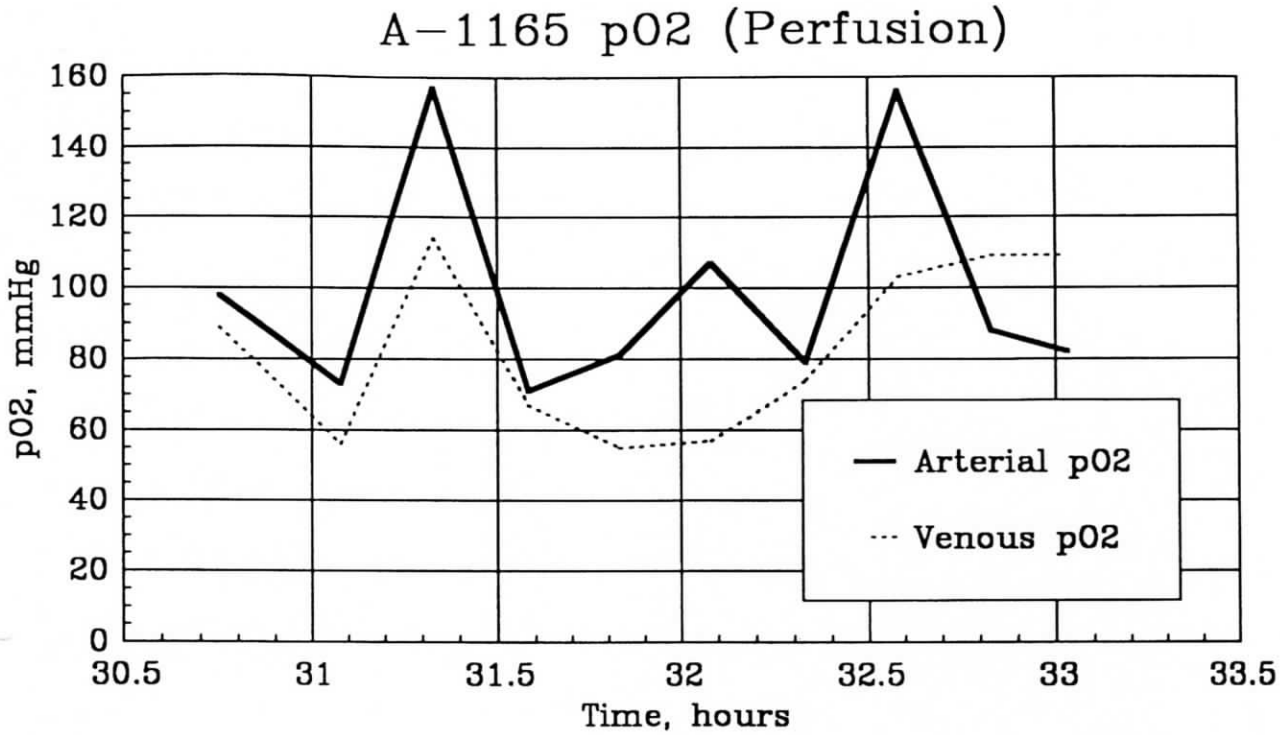
A periosteal elevator was used to expose the bone approximately 3 cm to the right of the midline. A 10 mm hole was made with a neuro burr and drill. The dura mater was opened and trimmed away with iris scissors to expose approximately 6 to 8 mm of the cortical surface. The burr hole was opened at 21:10; the pial vessels were noted to be free of blood and the cortical surface bulged approximately 1mm to 2 mm into the burr hole.

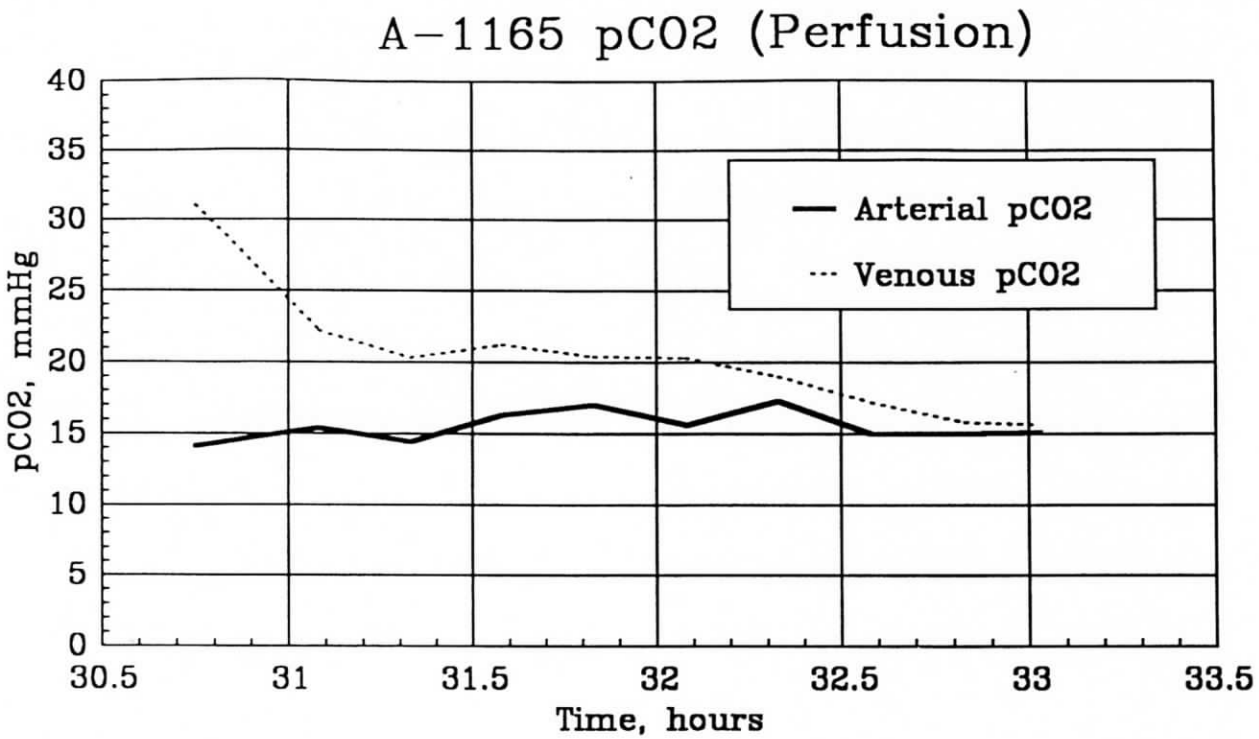
At 22:39 the burr hole was filled with bone wax and the scalp closed with surgical staples. Owing to economic considerations no temperature probe was placed on the cortical surface. Temperature descent to -77°C was monitored with a pharyngeal probe in addition to an external probe placed at the right temple on the surface of the head. These probes were Instrument Laboratories 53-20-507, "load type", 20 gauge, teflon-coated copper-constantan thermocouples. (The 53-20-507 TC probe was used to replace the clinical TC probe employed to monitor pharyngeal temperature during perfusion.) TC probes were anchored into place with surgical staples.

Blood Gas and pH Monitoring

Arterial and venous pO_2 and CO_2 determinations were first made at 20:10. Arterial pO_2 was 98 mmHg and arterial CO_2 was 14.1 mmHg and arterial pH was 7.91. Venous pO_2 was 89 mmHg, venous pCO_2 was 31 mmHg and venous pH was 7.17. Perfusion pH and gases were drawn again at 20:30 and were as follows: arterial pO_2 73 mmHg, arterial pCO_2 15.4 mmHg, arterial pH 7.68, venous pO_2 56, venous pCO_2 22.2, and venous pH 7.39. By 21:30 the arterial pressure had risen to 60 mm Hg and the arterial pressure was again reduced to 50 mm Hg by reducing the arterial flow rate to 400 cc/min. At that time arterial pH was 7.5, venous pH 7.43, pharyngeal temperature was 7.9°C, arterial temperature 6.8°C, sinus temperature was 7.0°C. A final determination of arterial and venous pO_2 and pCO_2 was made at 22:27 and were as follows: arterial pO_2 82 mmHg, arterial pCO_2 15.1, arterial pH 7.61, venous pO_2 109, venous pCO_2 15.6, and venous pH 7.73. Perfusion pH, pO_2, and pCO_2 are presented graphically below.




A−1165 pO2 (Perfusion)
pO2, mmHg
160
140
120
100
80
60
40
20
0
30.5
31
31.5
32
32.5
33
33.5
Time, hours
Arterial pO2
Venous pO2


A−1165 pCO2 (Perfusion)
pCO2, mmHg
40
35
30
25
20
15
10
5
0
30.5
31
31.5
32
32.5
33
33.5
Time, hours
Arterial pCO2
Venous pCO2

Cephalic Isolation

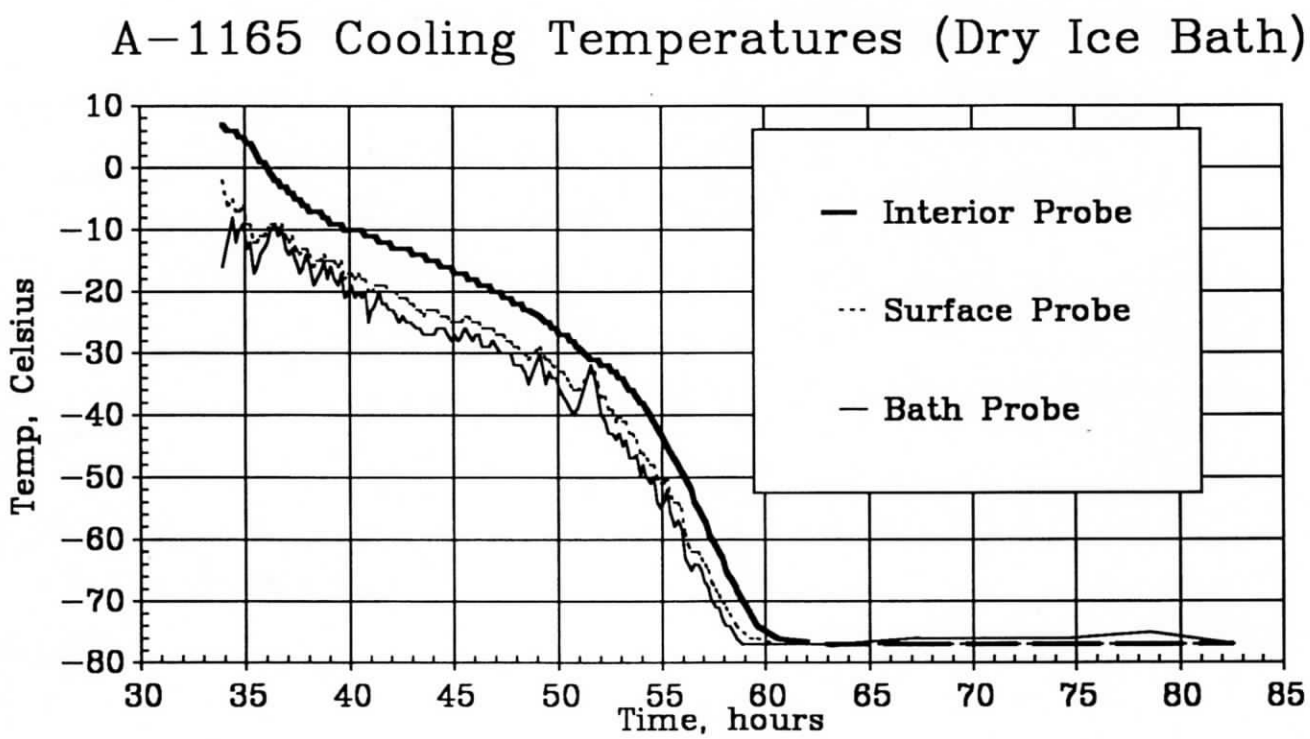
Surgery for cephalic isolation was begun at 23:00 with a circumferential skin incision made at the base of the neck and extended anteriorly and posteriorly to just below the margins of the clavicle. The skin was dissected free from the underlying connective tissue up to the level of the 5th cervical vertebrae to form skin flaps. The cervical musculature and other anatomical structures were then severed with a #10 scalpel blade down to the junction of the 5th and 6th cervical vertebrae. A Gigli saw was then passed under the vertebral column and the cut was made at approximately the level of the 5th cervical vertebrae, which freed the head from the body.

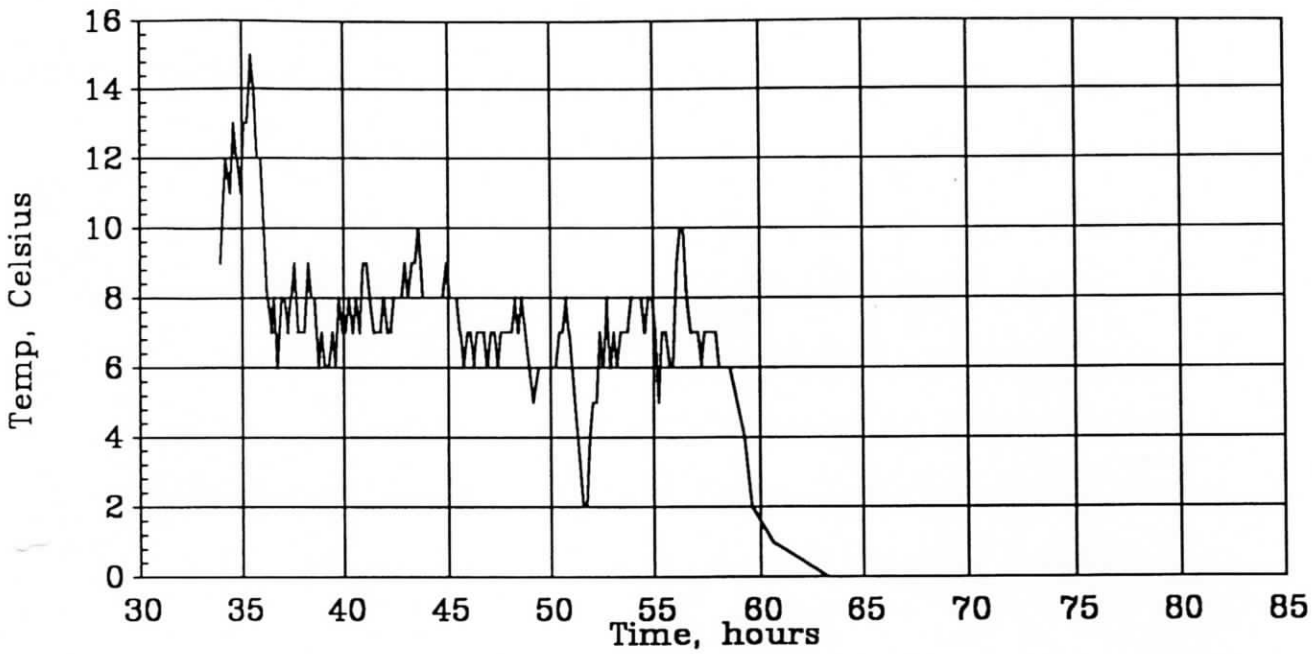
The cervical skin and musculature were observed to be dark in color, evenly stiff, and somewhat waxy in texture, consistent with uniform glycerolization.

Skin flaps were then closed over the stump of the neck using a skin stapler, after the edges of the flaps were first approximated using interrupted 2-0 Tycron suture. Cephalic isolation was completed at 23:15. Weight of the cephalon was 3.9 kg.

Cooling to Dry Ice Temperature

The patient (cephalon) was placed inside two 2 mil polyethylene plastic bags and lowered into a 15 liter bath of 5 centistoke polydimethylsiloxane oil (Silcool) which had been pre-cooled to -16°C. The first temperature readings taken at 23:21 were: pharyngeal, 7.0°C; surface, -2.0°C; and bath, -16.0°C. Cooling to -77°C began at a rate of approximately -1.8°C per hour and ended at a rate of approximately -6.5°C per hour. The bath to pharyngeal temperature differential was approximately 10°C to -40°C and 5°C to 7°C from -40°C to -77°C. Cooling to -77°C was complete by 02:00 on 10/10/88. The patient's dry ice cooling curve and surface to core temperature differential is presented graphically below. Time shown is in decimal hours post-arrest.

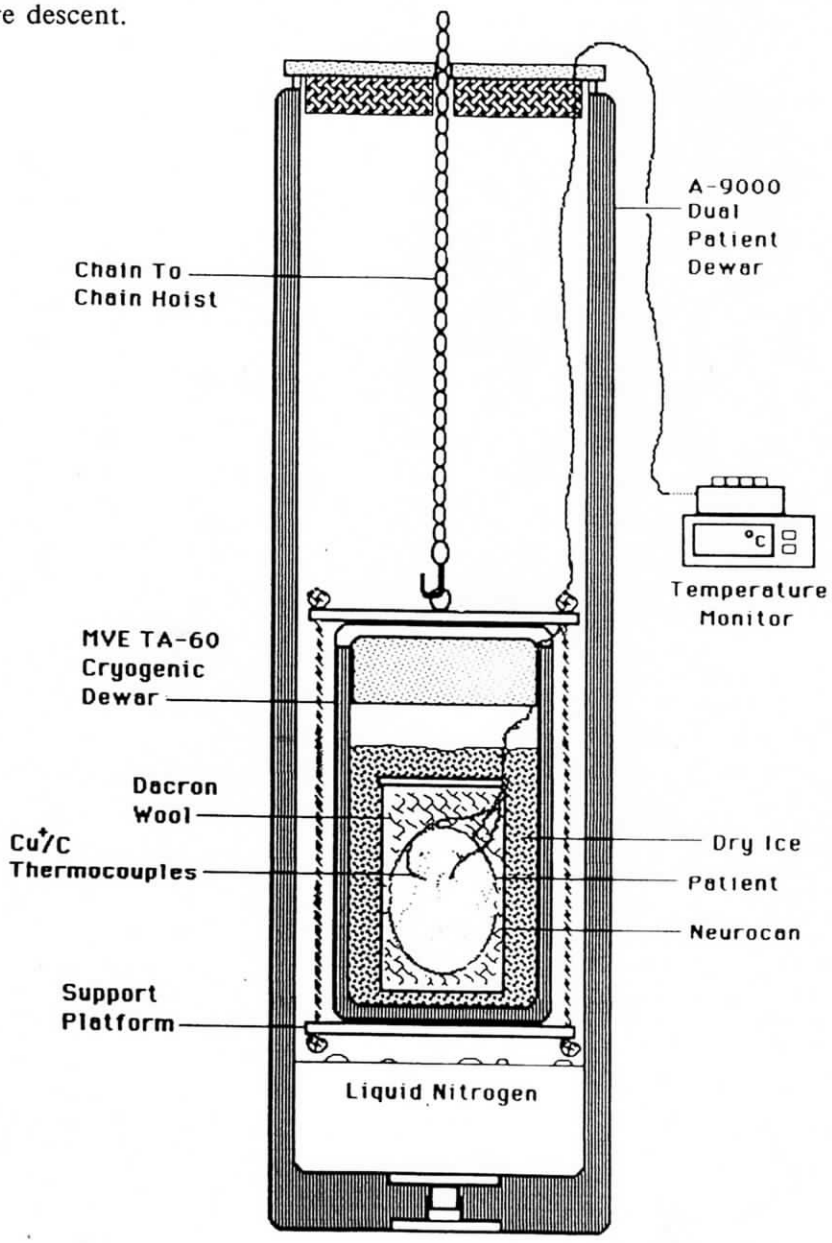



A-1165 Surface-Core Difference (Dry Ice Bath)
Temp, Celsius
16
14
12
10
8
6
4
2
0
30
35
40
45
50
55
60
65
70
75
80
85
Time, hours

Cooling to -196°C

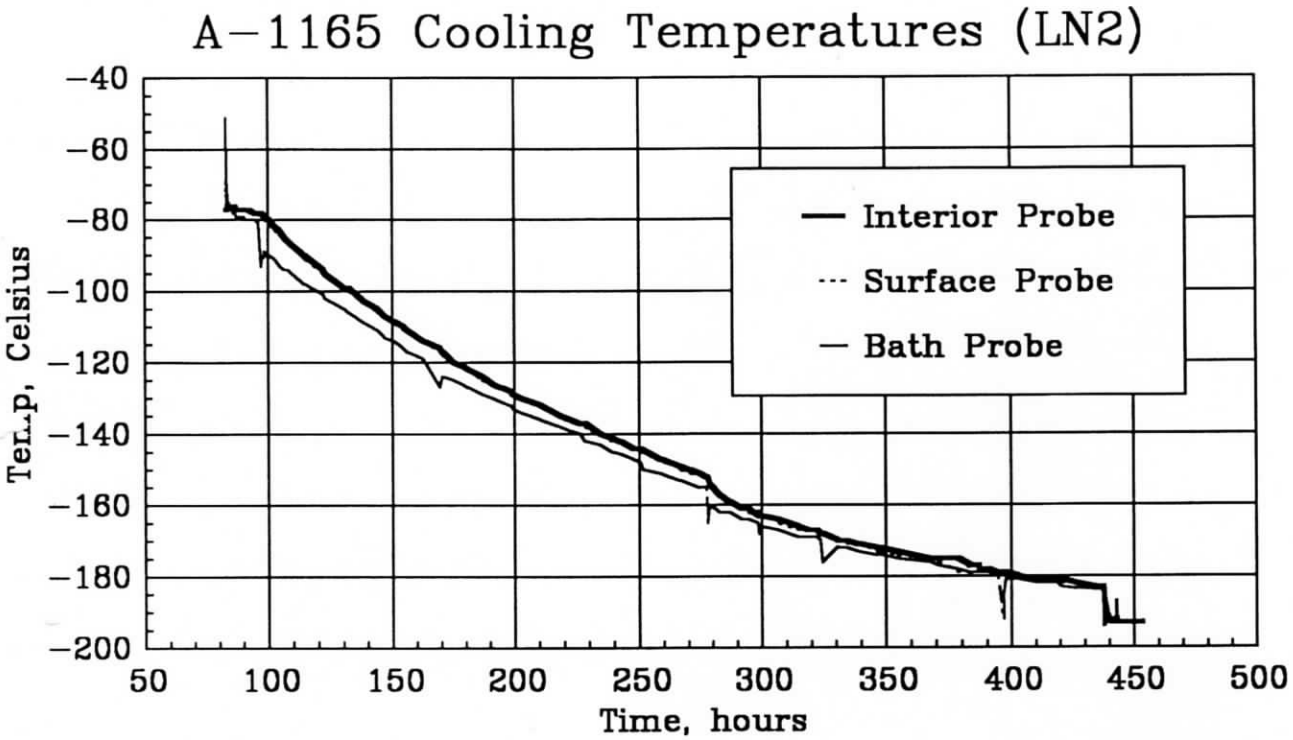
On 10/11/88 the patient was removed from the Silcool bath, the outer Silcool-wetted plastic bag was stripped off, and the patient was placed inside a polyester pillow case resting on a bed of Dacron wool at the bottom of an aluminum neurocan. This entire assembly had been pre-cooled by being nested inside a Linde LR-40 cryogenic dewar with the annulus between the neurocan and the dewar being filled with dry ice (see accompanying diagram). The pillow case was then closed with a white cotton shoe lace to which was affixed a stainless steel tag identifying the patient, and the lid of the LR-40 was closed with the TC probes externalized through the top.

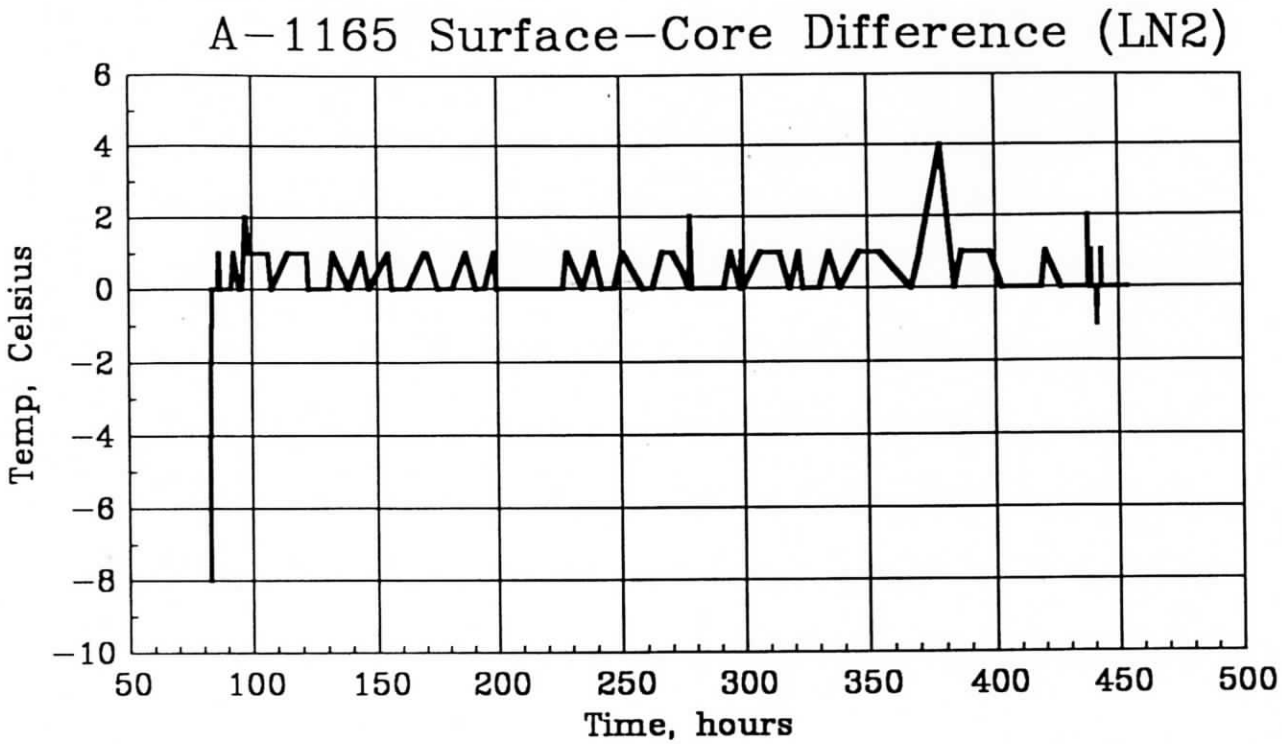
The LR-40 was then placed on a support platform of 3/4" plywood which could be lowered or raised on a chain hoist. The LR-40 was lowered into a 380 liter custom manufactured cryogenic dewar (CD) with internal measurements of 22" in diameter x 61" deep to which 160 liters of liquid nitrogen had been added. The LR-40 was lowered until the bottom 2" was submerged in liquid nitrogen and a slotted plywood, fiberglass insulated lid put in place. Thermocouple probes were led out of the CD and connected to a Omega 2165A thermocouple thermometer. The LR-40 was then raised or lowered as needed to obtain the desired rate of temperature descent.



Liquid Nitrogen Cooling Assembly

The initial rate of descent was approximately -0.42°C per hour and the final rate of descent was approximately -0.14°C per hour. Cooling to -196°C was achieved at 00:16 on 10/26/88, at which time the patient was placed into long-term cryogenic storage by submersion in liquid nitrogen in an MVE A-8000 cryogenic dewar. The patient's liquid nitrogen cooling curve and surface to core temperature differential is presented graphically below.





Laboratory Evaluations

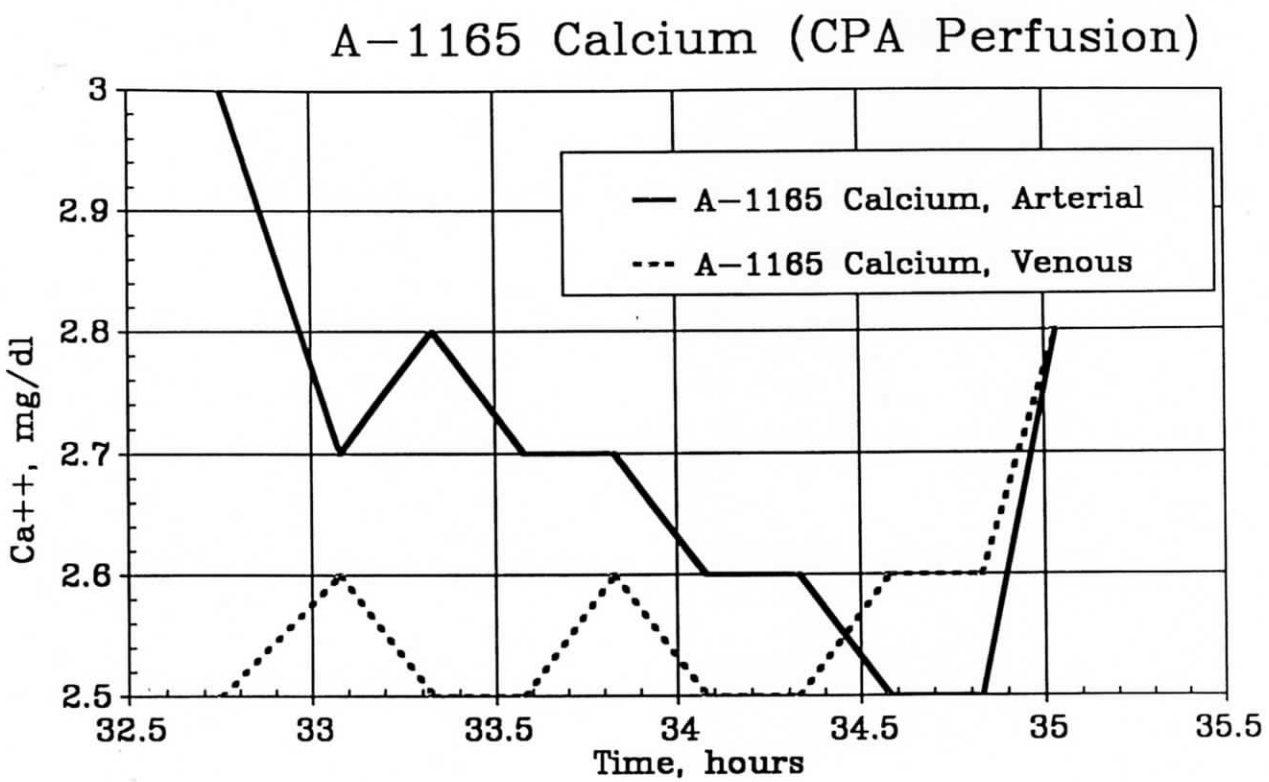
Laboratory evaluations of samples taken during cryoprotective perfusion are presented in full in both graphic and tabular form as an addendum to this document. The following general observations are made:

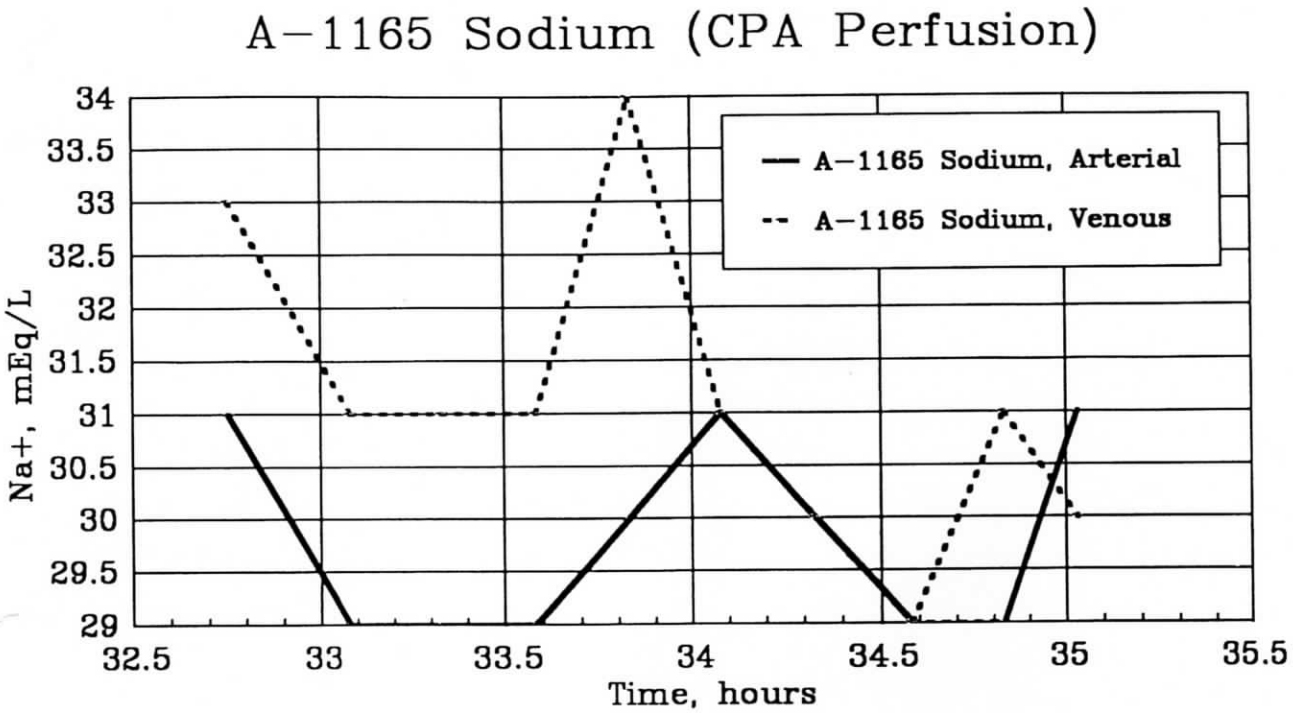
All serum and venous effluent submitted for analysis was unhemolyzed and normal in appearance.

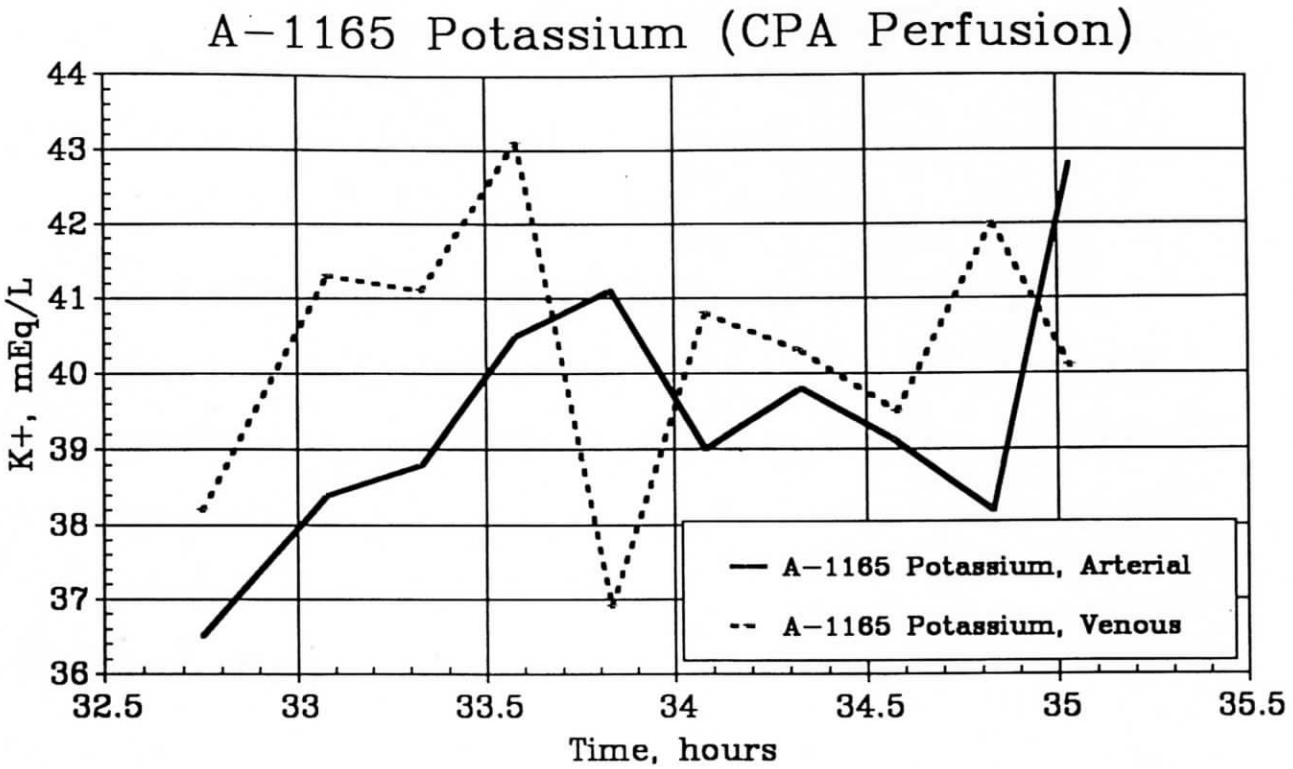
LDH, SGOT and SGPT were significantly elevated over "baseline" determinations made approximately 24-hours pre-deanimation, probably as a result of both the lengthy agonal period of shock and the likely poor perfusion during HLR-supported transport. Somewhat surprizingly the alkaline phosphatase levels were not elevated over pre-deanimation levels and were in fact markedly lower.

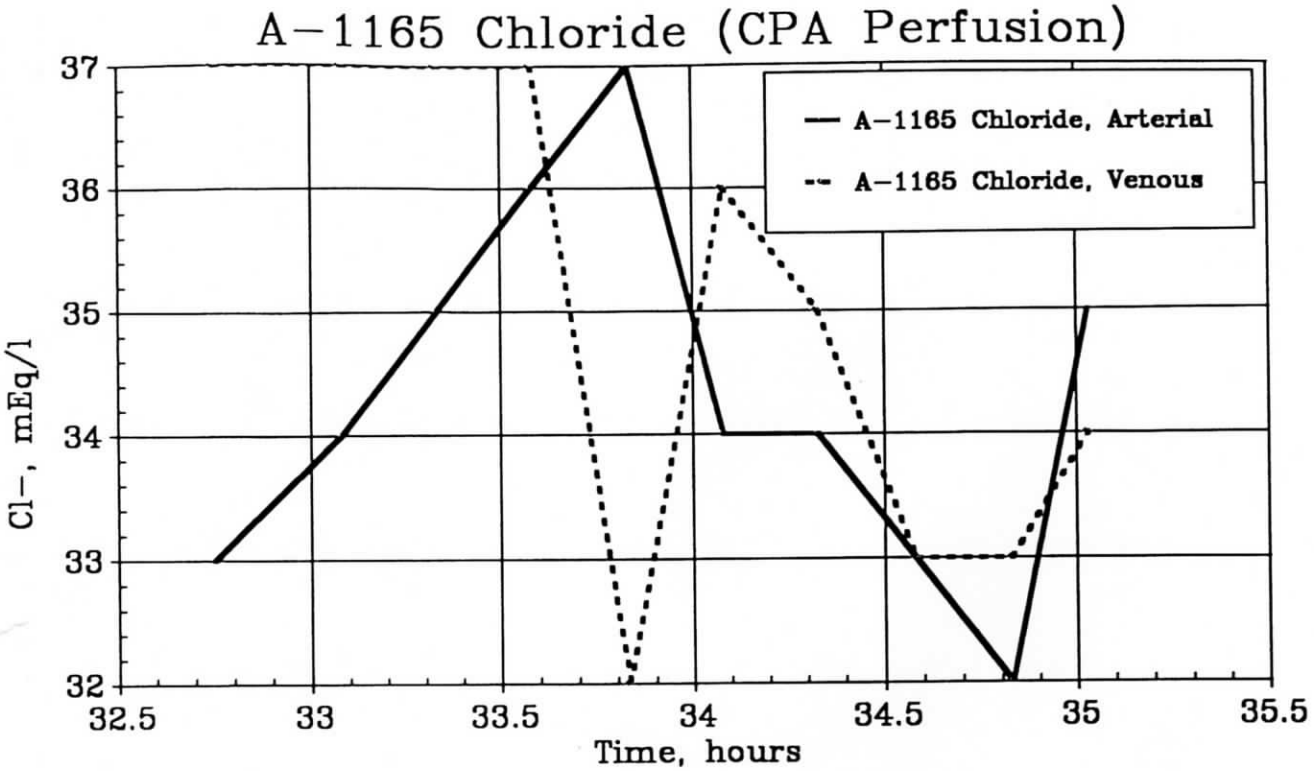
Predictably tissue specific enzyme levels declined during TBW, rose again during the cold ischemic period of air transport, and then declined to more or less equilibrium values during cryoprotective perfusion indicating ongoing release from tissues.

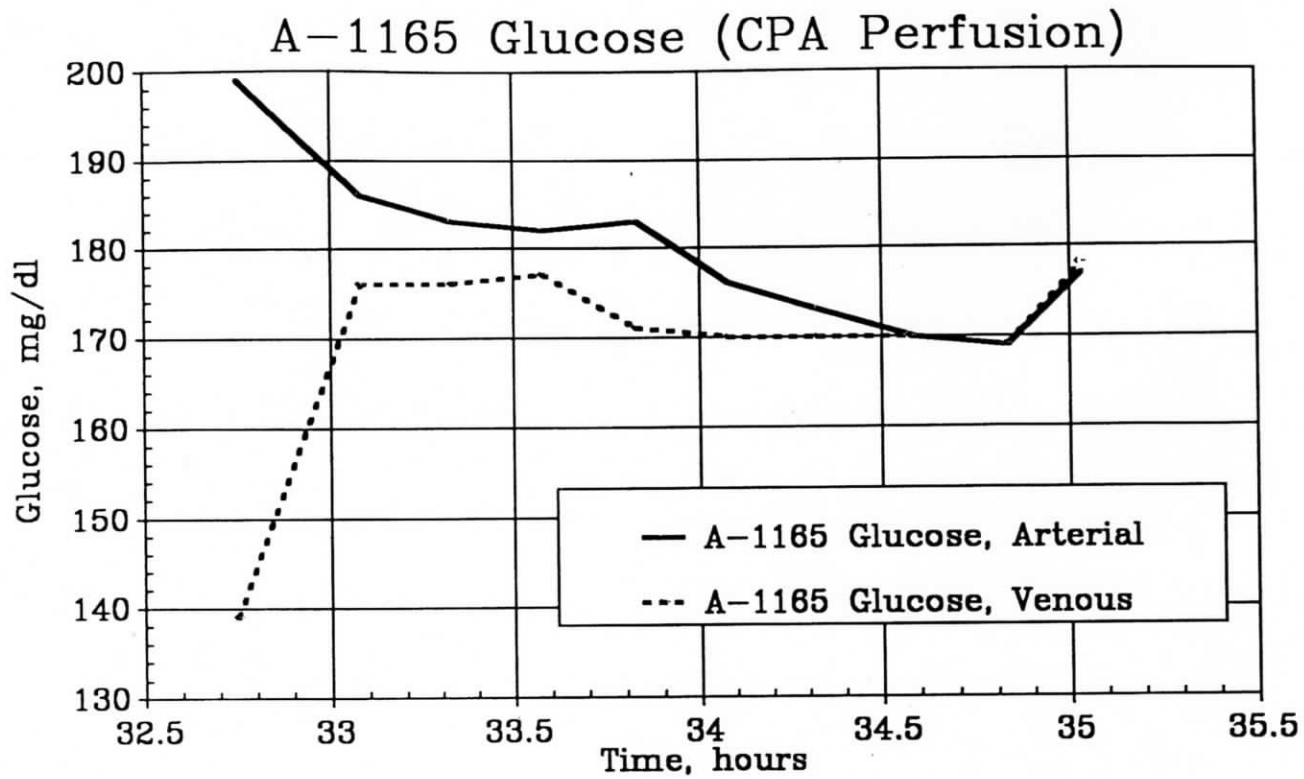
The initial blood glucose reading taken near the end of HLR-support was 19 mg/dl, probably indicative of grossly inadequate hepatic perfusion and probably grossly inadequate systemic perfusion as well.

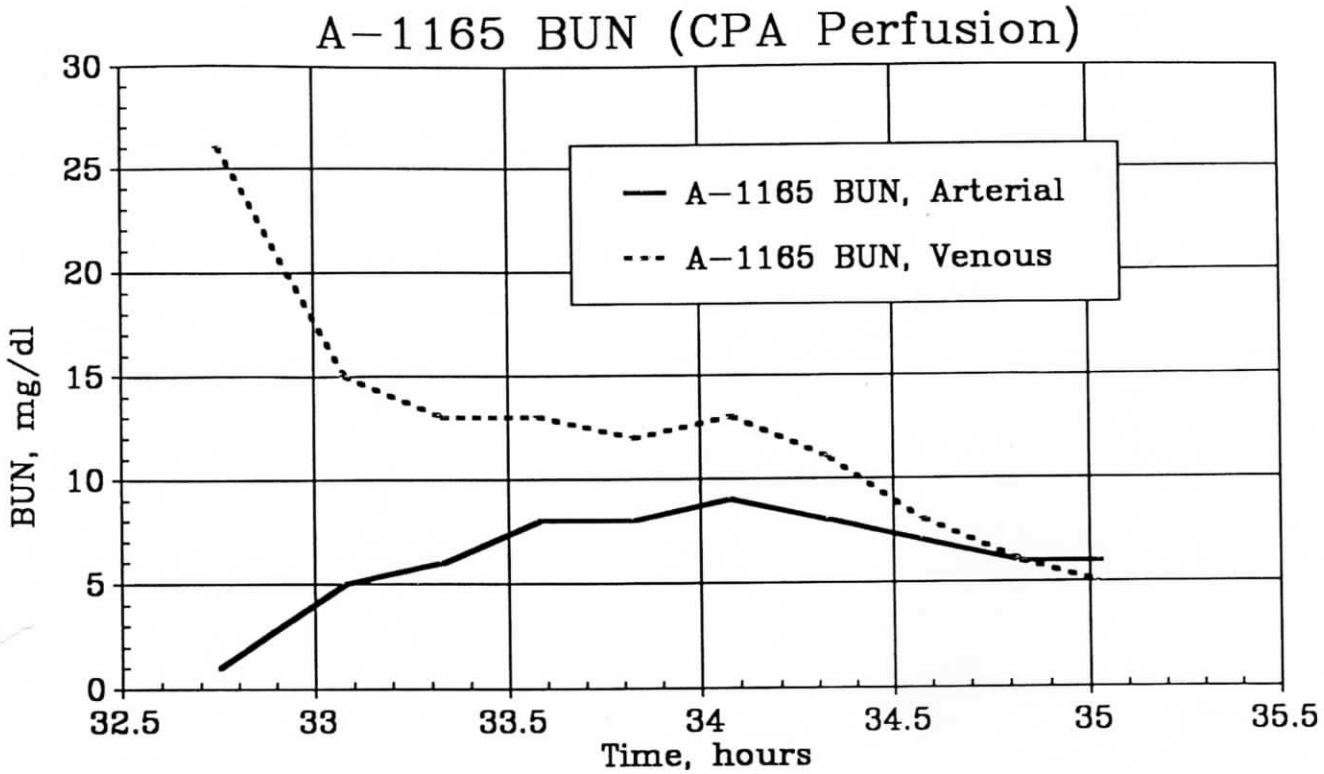


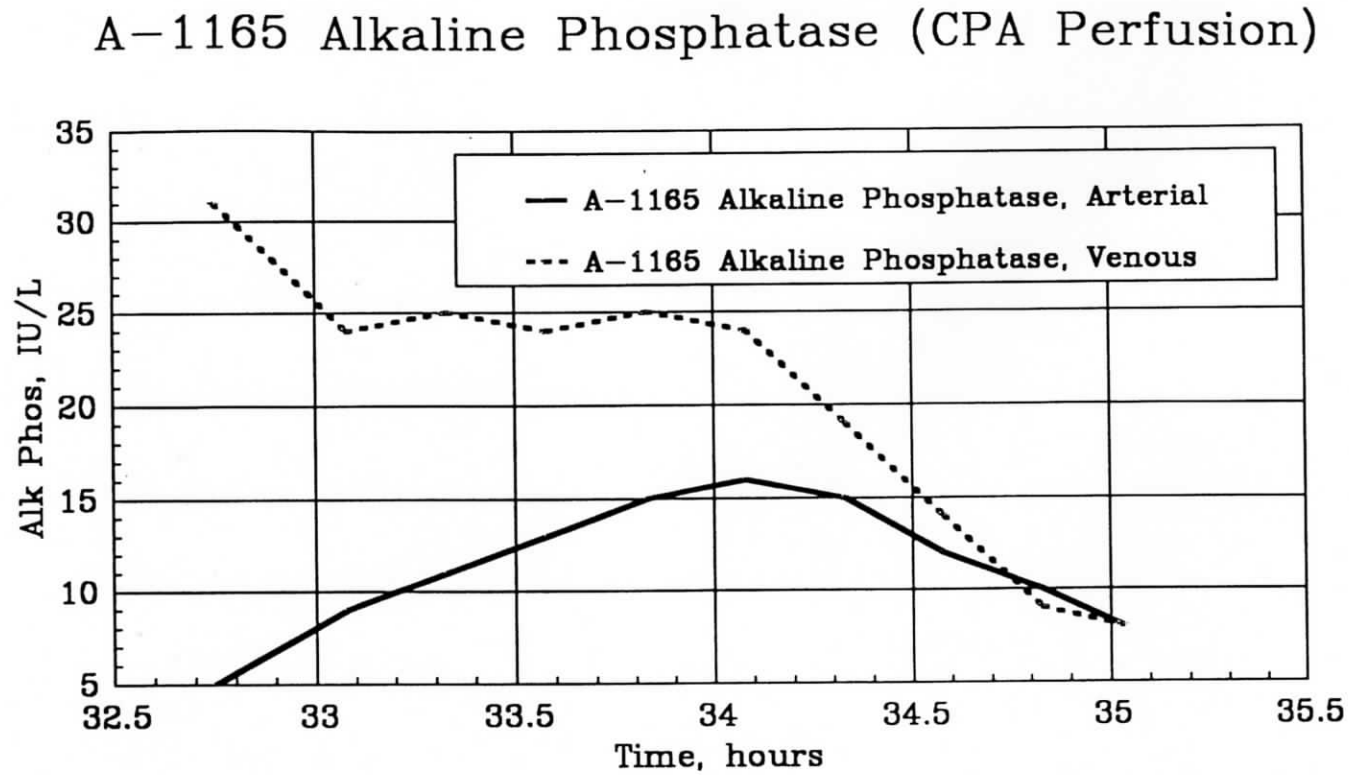

A−1165 Sodium (CPA Perfusion)
Na+, mEq/L
34
33.5
33
32.5
32
31.5
31
30.5
30
29.5
29
32.5
33
33.5
34
34.5
35
35.5
Time, hours
A−1165 Sodium, Arterial
A−1165 Sodium, Venous

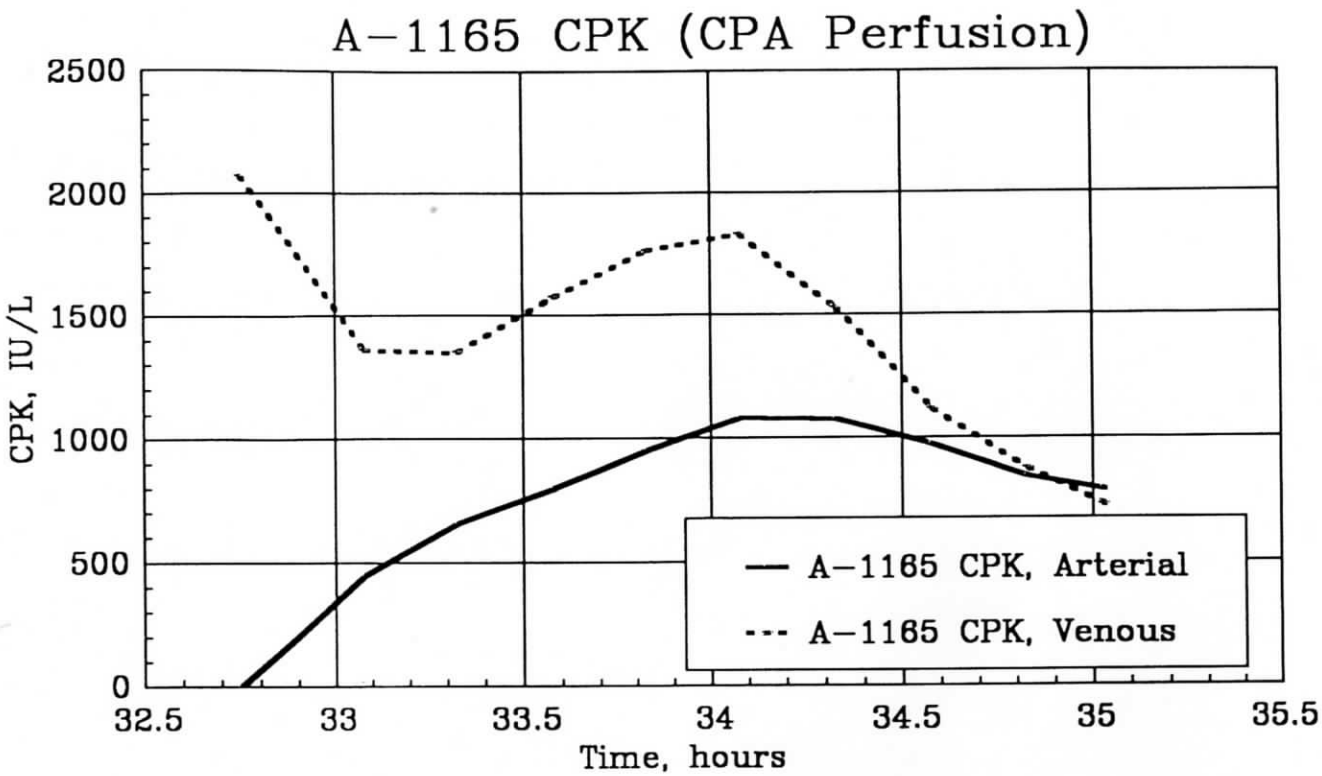

A−1165 Potassium (CPA Perfusion)
K+, mEq/L
44
43
42
41
40
39
38
37
36
32.5
33
33.5
34
34.5
35
35.5
Time, hours
A−1165 Potassium, Arterial
A−1165 Potassium, Venous

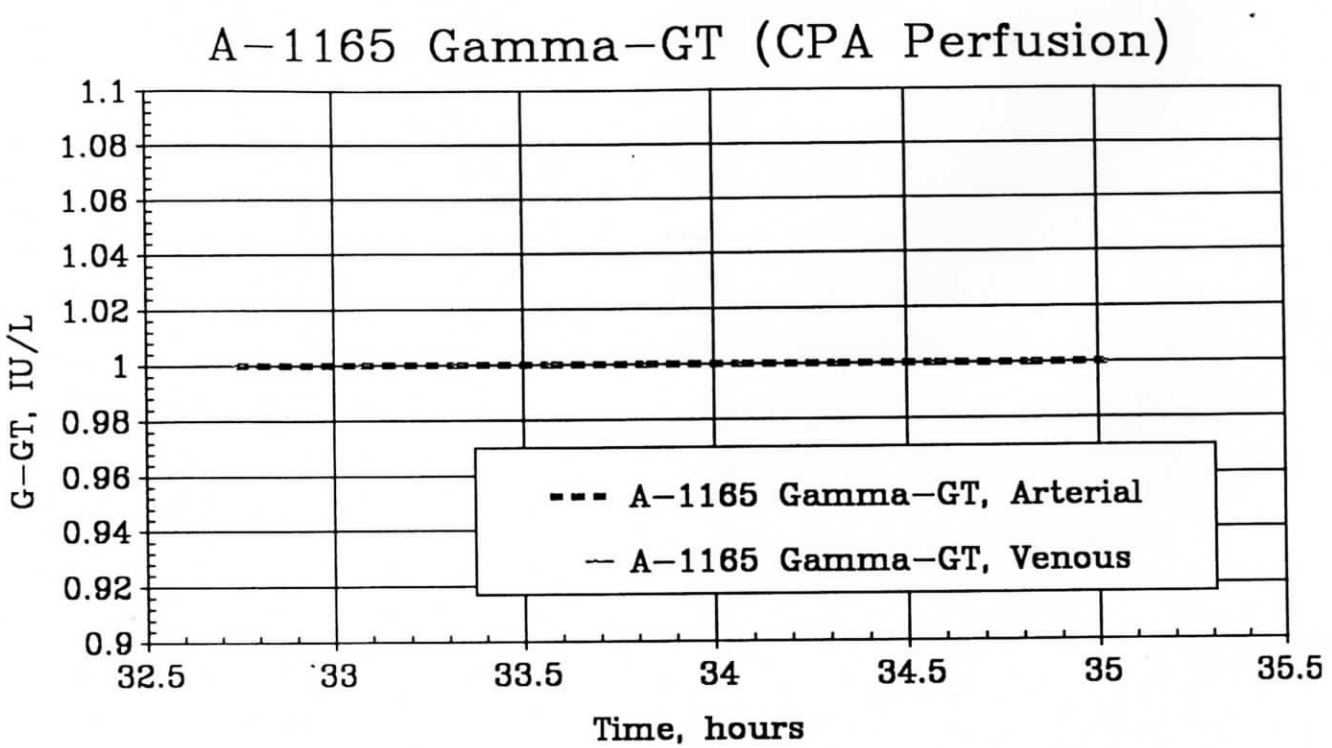

A−1165 Chloride (CPA Perfusion)
A−1165 Chloride, Arterial
A−1165 Chloride, Venous
Cl−, mEq/l
37
36
35
34
33
32
32.5
33
33.5
34
34.5
35
35.5
Time, hours

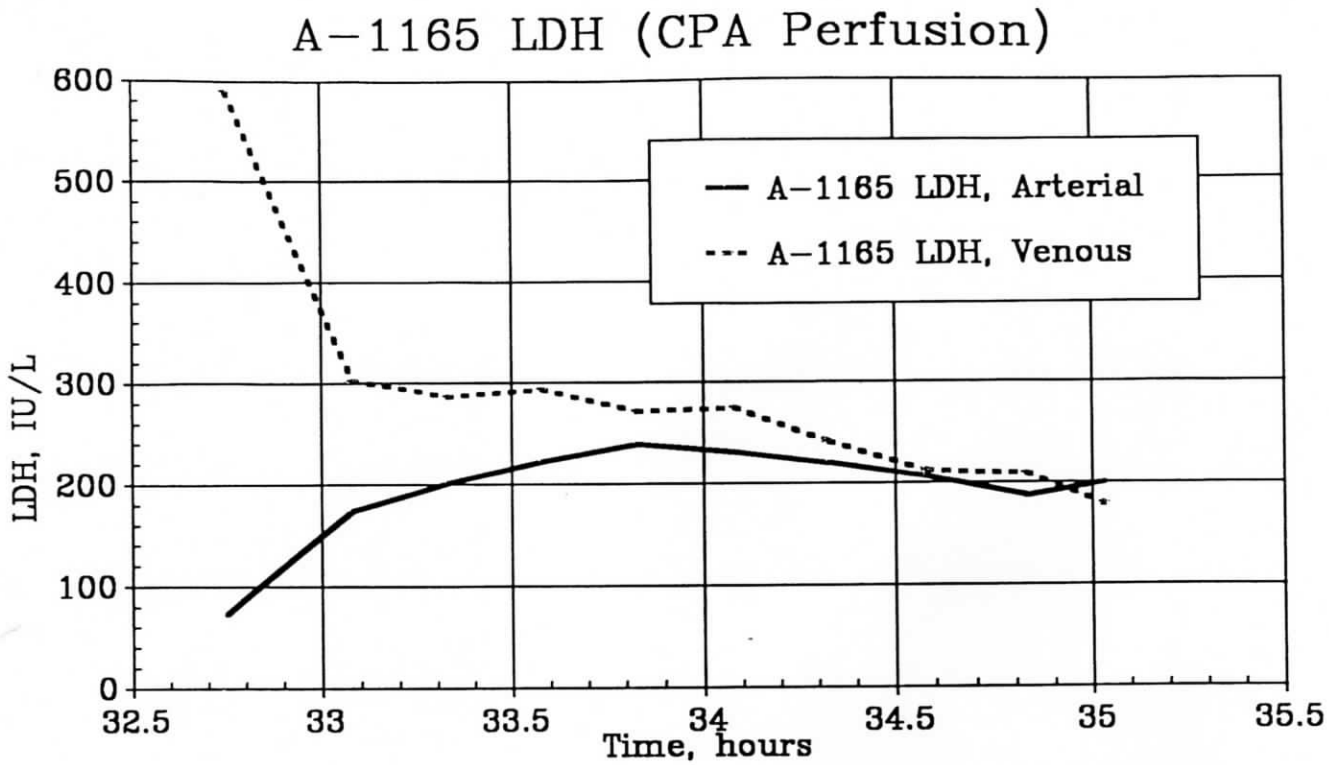

A−1165 Glucose (CPA Perfusion)
A−1165 Glucose, Arterial
A−1165 Glucose, Venous
Glucose, mg/dl
200
190
180
170
160
150
140
130
32.5
33
33.5
34
34.5
35
35.5
Time, hours

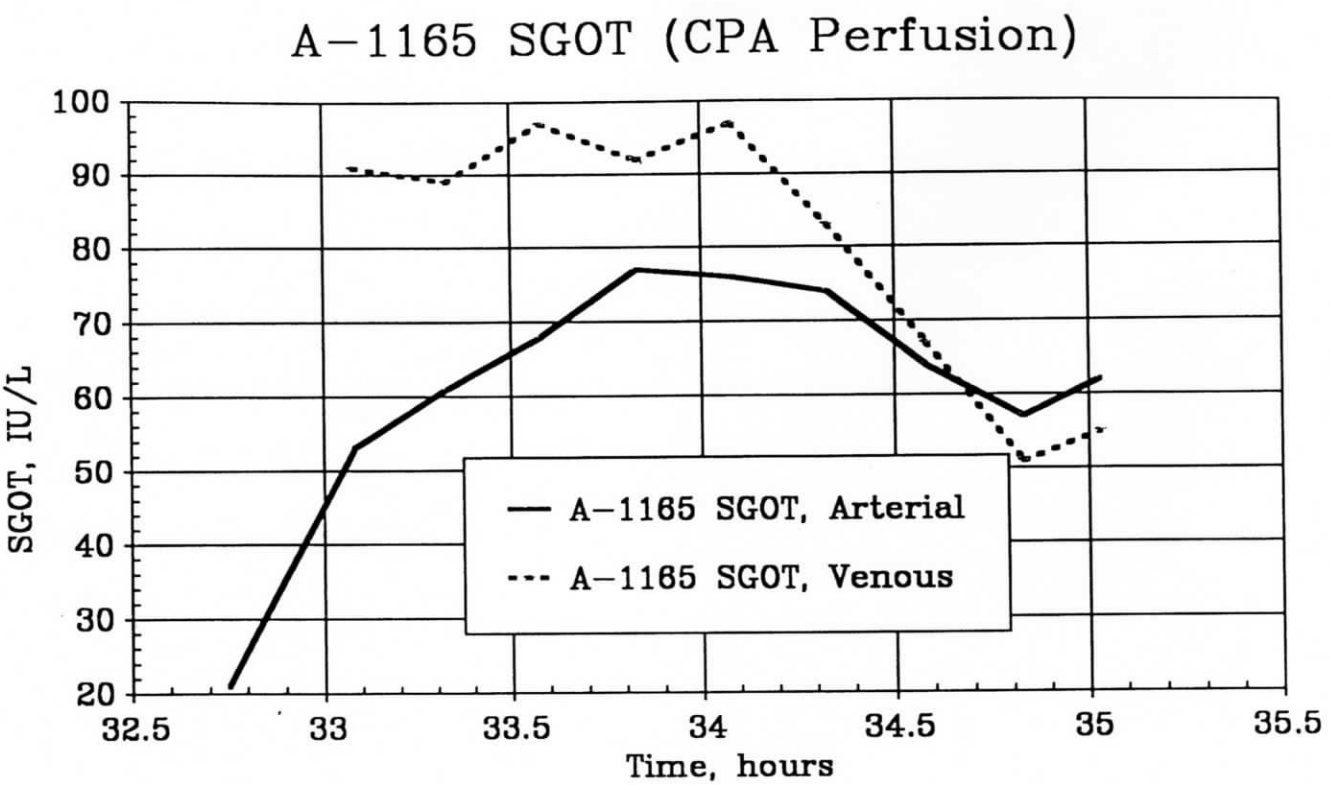

A–1165 BUN (CPA Perfusion)
A–1165 BUN, Arterial
A–1165 BUN, Venous
BUN, mg/dl
Time, hours
0
5
10
15
20
25
30
32.5
33
33.5
34
34.5
35
35.5

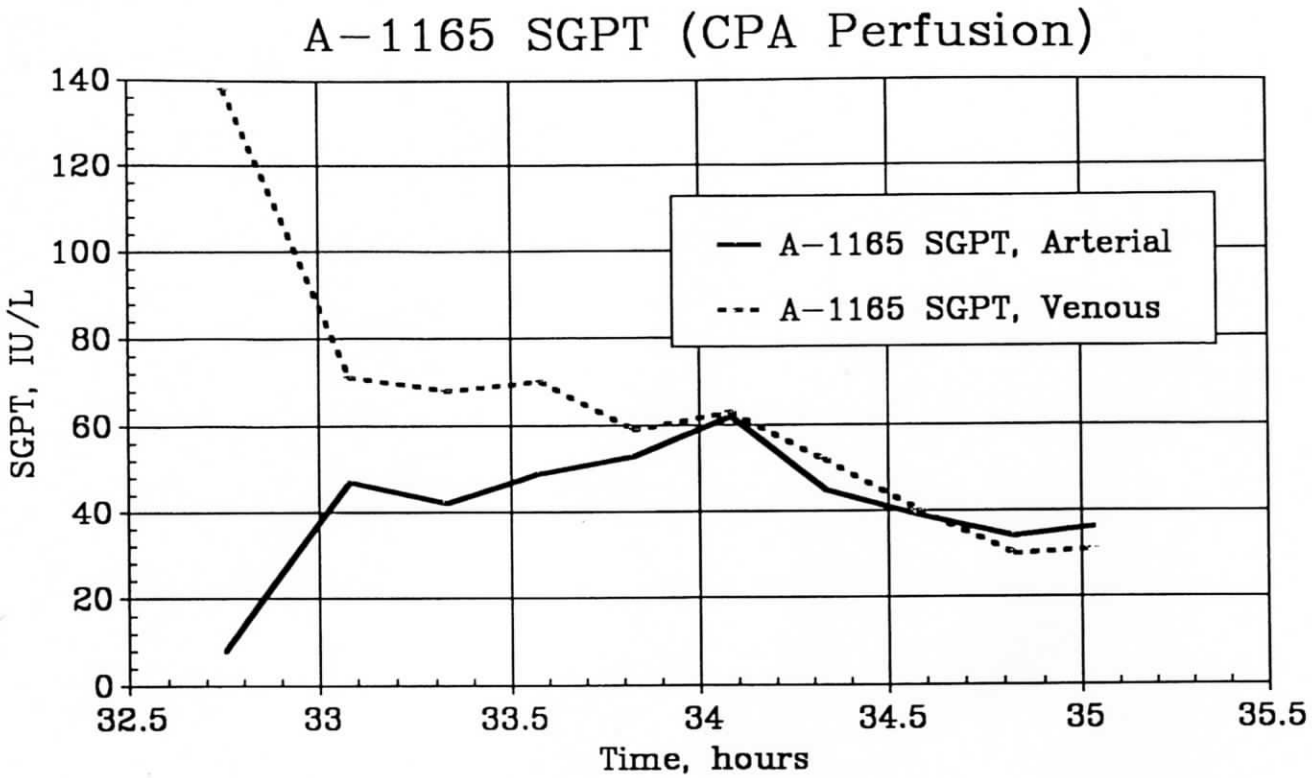

A–1165 Alkaline Phosphatase (CPA Perfusion)
A–1165 Alkaline Phosphatase, Arterial
A–1165 Alkaline Phosphatase, Venous
Alk Phos, IU/L
Time, hours
5
10
15
20
25
30
35
32.5
33
33.5
34
34.5
35
35.5


A−1165 CPK (CPA Perfusion)
CPK, IU/L
2500
2000
1500
1000
500
0
32.5
33
33.5
34
34.5
35
35.5
Time, hours
A−1165 CPK, Arterial
A−1165 CPK, Venous


A−1165 Gamma−GT (CPA Perfusion)
G−GT, IU/L
1.1
1.08
1.06
1.04
1.02
1
0.98
0.96
0.94
0.92
0.9
32.5
33
33.5
34
34.5
35
35.5
Time, hours
A−1165 Gamma−GT, Arterial
A−1165 Gamma−GT, Venous


A-1165 LDH (CPA Perfusion)
600
500
400
300
200
100
0
LDH, IU/L
32.5
33
33.5
34
34.5
35
35.5
Time, hours
A-1165 LDH, Arterial
A-1165 LDH, Venous


A-1165 SGOT (CPA Perfusion)
100
90
80
70
60
50
40
30
20
SGOT, IU/L
32.5
33
33.5
34
34.5
35
35.5
Time, hours
A-1165 SGOT, Arterial
A-1165 SGOT, Venous


A–1165 SGPT (CPA Perfusion)
140
120
100
80
60
40
20
0
SGPT, IU/L
32.5
33
33.5
34
34.5
35
35.5
Time, hours
A–1165 SGPT, Arterial
A–1165 SGPT, Venous