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Feasibility of a Porcine Adult Intensive Care Model

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A porcine adult ICU model would be useful for several avenues of investigation relevant to the care of critically ill patients. The purpose of the experiments reported here was to test the feasibility of such a model, using healthy swine. Swine (n = 4; body weight, 76 ± 5 kg) were instrumented with endotracheal, bladder, and central arterial and venous catheters, and were admitted to the intensive care unit (ICU) while undergoing mechanical ventilation under the continuous care of nurses. Cardiopulmonary parameters were monitored continuously, and serum biochemical parameters were measured intermittently. Survival was seven days in subject 1 and five and a half days in subject 2. Subjects 3 and 4 survived an abbreviated protocol (44 and 41 h, respectively). Care of the subjects was complicated by iatrogenic hemorrhage (n = 3), pneumonia (n = 2), and acute respiratory distress syndrome (n = 1). One subject was free of complications. Critically ill swine ≥ 70 kg can survive mechanical ventilation in the ICU for up to seven days. When iatrogenic injury occurs, swine respond well to clinical care protocols. Further testing is needed to develop a reproducible model and determine whether healthy swine can survive the ICU environment for longer than 41 h.

The mechanisms of pathophysiologic problems in the critically ill are difficult to study because of the interactive effects of treatment, pre-existing and concomitant disease, and the environment of the intensive care unit (ICU). Because neither the internal milieu nor the external environment of critically ill patients can be controlled, little data exist on the time structures of biophysiologic function during critical illness. Our long-term objective is to shorten the duration of mechanical ventilation in adult critically ill patients by timing ventilator withdrawal (weaning) according to optimal phases of biophysiologic time structures (e.g., circadian rhythms). One challenge is to separate the effects of the ICU environment from the effects of patient illness on biorhythms during mechanical ventilation and weaning. To address this challenge, we attempted to develop a novel experimental ICU model that could permit study of healthy and critically ill porcine subjects.

The purpose of the experiments reported here was to test the feasibility of a porcine adult ICU model for use in future work to test hypotheses about the effects of mechanical ventilation and weaning on biophysiologic time structures. The American Association of Critical-Care Nurses' National Study Group on Weaning from Mechanical Ventilation defined long-term mechanical ventilation as that lasting longer than three days (9, 16). Long-term mechanically ventilated patients typically have difficulty with weaning, and this group of patients would benefit most from the identification of optimal times for weaning. Therefore, we require an experimental model that can be maintained under ICU conditions for longer than three days.

Few experimental models have been described that can be generalized to the adult ICU. Gronert and colleagues (6, 7) described a canine ICU model. They kept healthy beagles weighing 9 to 17 kg in an experimental ICU on mechanical ventilation for up to three weeks. However, our interest is in weaning critically ill adults, and a large animal model would better represent the adult ICU patient and environment by allowing use of equipment, drugs, fluids, and protocols used to care for adult patients. The domestic farm pig was selected because it has circadian rhythms in core body temperature and cortisol concentration (2, 5, 8, 17), marker rhythms for circadian time structure, that are similar to those of humans. We hypothesized that healthy pigs can survive mechanical ventilation, weaning, and the ICU environment by use of good nursing care and prevention of complications.

Under anesthesia and aseptic conditions, subjects were instrumented surgically with indwelling catheters. After instrumentation, they were placed in hospital beds and transferred from the operating room to the experimental porcine ICU. The full model was tested on two subjects (Yorkshire/Blue Butt cross) from an open herd (Russell Barham, Midway, Tex.). We encountered problems with the arterial catheter, anticoagulation, and farm-acquired pneumonia. Thereafter, an abbreviated model was tested on two subjects (Yorkshire/Duroc cross) from a closed herd (HDH Swine Farm, Boerne, Tex.), using different arterial catheters and a revised anticoagulation protocol. Because of resource constraints, the abbreviated model was scheduled for 40 h.

Subjects from the closed herd were vaccinated against atrophic rhinitis associated with *Bordetella bronchiseptica*, erysipelas caused by *Erysipelothrix rhusiopathiae*, and *Pasteurella multocida* infection at weaning (five weeks of age). Since 1980, the vendor has introduced all blood lines through artificial insemination, thereby maintaining a closed herd, and all breeding animals have tested normal for malignant hyperthermia. On admission to the research facility (17 weeks), the subjects were dewormed and given oxytetracycline.

Materials and Methods

Animals. The University of Texas Health Science Center at

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Houston and the Texas Heart Institute institutional animal care and use committees approved these non-survival experiments. Care, handling, and procedures conformed to the US Public Health Service guidelines for humane care and use of laboratory animals (20, 21).

The sample was four male, domestic farm pigs weighing 69 to 80 kg. Subjects were transported in pairs from a farm to the research facility. The first pair of subjects was composed of littermates, and all subjects were four to five months old at the time of transfer. As is customary at the animal care facility, the subjects were housed singly in pens that were situated side by side, and pigs were acclimated for 22 to 27 days. The second pair was housed in an isolation room; access was limited, and visitors wore protective garb while in the room. Daily routine during conditioning consisted of one feeding of Lab Porcine Diet Grower (LabDiet, Formula 5084, PMI Nutrition International, Inc., Brentwood, Mo.), water ad libitum, exercise, and visits from members of the research team and animal husbandry staff. The housing room was dark, except for a nightlight, between 7 p.m. and 7 a.m. and was lighted between 7 a.m. and 7 p.m. by use of an automatic room timer.

Instrumentation. Food was withheld for 18 to 24 h, and each pig was bathed before the experiment. After sedation by intramuscular administration of atropine sulfate (0.05 mg/kg of body weight), acepromazine (0.11 to 0.25 mg/kg), and ketamine HCl (20 mg/kg), the subject was intubated with a 9.0- to 10.0-mm cuffed endotracheal tube (Aire-Cuf, Bivona Medical Technologies, Inc. Gary, Ind.). Electrocardiogram leads were applied to monitor heart rate and rhythm, a bite block was inserted orally and secured to the mandible, and a whole-body betadine scrub was done.

Anesthesia was maintained with 1.0 to 3.0% isoflurane for survival surgery. The animal's right internal jugular vein was cannulated by use of an 8.5-F introducer. An 8-F, 6 lumen (i.e., ports), flow-directed thermodilution fiberoptic continuous cardiac output pulmonary artery catheter (OptiQ, Abbott Critical Care Systems, North Chicago, Ill.) was inserted through an introducer into the pulmonary artery by use of waveform observation. A 17-mm internal diameter polyvinyl catheter, measuring 10 cm in length, was inserted into the right carotid and right femoral artery of subjects 1 and 2; the length of the catheter was constrained for later insertion of an intra-arterial sensor for continuous blood gas monitoring. In subjects 3 and 4, a new intra-arterial blood gas sensor (Neotrend, Model N7004S, Diametrics Medical, Inc.) was used with an umbilical artery catheter that was 27.5 cm long. The right femoral vein was cannulated and used solely for infusion of parenteral nutrition by pump (Gemini PC-2T volumetric pump/controller, IMED Corporation, San Diego, Calif.). All major vessels were accessed by cut down. A YSI Series 400 thermistor embedded in a 16-F urinary bladder drainage catheter (Mon-a-therm Foley-Temp, Mallinckrodt Medical, Inc., St. Louis, Mo.) was surgically tied into the bladder, using the suprapubic approach described elsewhere (13).

After instrumentation, a bolus of heparin (1,000 U/kg) was administered intravenously to the subject. Anesthesia was discontinued, the subject was transferred to a hospital bed (Advance 2000 series, Hill-Rom, Batesville, Ind.), and was admitted to the experimental ICU.

Porcine ICU. An operating room in the research laboratory served as the porcine ICU. Full-room lighting was on from 7 a.m. to 7 p.m. From 7 p.m. to 7 a.m., the overhead surgical lights were on at 50% capacity. Room temperature varied from 22 to 28°C, and relative humidity ranged between 15 and 42%. Universal precautions were in effect throughout the experiments. Senior nurse clinicians and nursing students cared for the subjects in 12-h shifts. The first two pigs were studied simultaneously, with two to three caregivers continuously present for care, monitoring, and data collection. Author (SKH) and the clinical veterinarian (Gil Costas, DVM) were either present or available on a 24-h basis for consultation. The last two pigs were studied individually, with author (SKH) and a graduate nursing student providing round-the-clock care.

Subjects were placed on a volume-controlled mechanical ventilator (Puritan Bennett Series 7200 or PB840, Mallinckrodt Medical, Inc.). Ports of the pulmonary artery catheter were used for continuous cardiac output monitoring (QVue CCO Monitoring System, Abbott Laboratories, North Chicago, Ill.), transduced pressure monitoring (Universal quartz pressure transducer, Model 1290C, Hewlett Packard, Andover, Mass.), and infusion of drugs and fluids. One arterial catheter provided blood sampling access and transduced pressure data through a physiologic monitor (SpaceLabs Medical, Inc., Redmond, Wash., or Eagle 4000 physiologic monitor, Marquette Electronics, Milwaukee, Wis.). A sensor was threaded through the ipsilateral arterial catheter and was connected to a continuous intra-arterial blood gas monitor (Paratrend 7, Diametrics Medical, Inc., St. Paul, Minn.). Electrocardiogram; pulse oximeter saturation (tongue or ear); mixed venous oxygen saturation; arterial, pulmonary artery, and central venous pressures; and bladder temperatures were monitored by use of the physiologic monitor. A 16-F clear polyvinyl orogastric tube was inserted to decompress the stomach with gravity drainage.

Light to moderate sedation was maintained by use of continuous intravenous infusion of pentobarbital sodium that was titrated to permit spontaneous respiration and movement without the subject chewing on the endotracheal tube. The infusion was started on admission to the porcine ICU at the rate of 5 mg/kg/h, and was adjusted for clinical effect by monitoring spontaneous muscular activity (e.g., extremity, head, and eye movements; coughing, swallowing), mandibular muscle tone, carpopedal reflex response to manual stimulation, shivering, spontaneous breathing rate, heart rate, mean arterial blood pressure, and $PaCO_2$. Buprenorphine was given intravenously in 0.1-mg doses as needed for alleviation of postoperative pain; the frequency varied on the basis of clinical signs of discomfort and cardiopulmonary parameters, but no subject received more than 0.8 mg in a 24-h period.

A clinical pharmacist and a swine veterinarian formulated the parenteral nutrition protocol. The nutritional objective was to maintain steady weight gain of 0.2 kg/d with protein accretion (4) and electrolyte balance. The daily regimen provided 37.4 kcal/kg/d, with 10% protein (Travasol 10% Amino Acid Solution, Baxter Healthcare Corp., Deerfield, Ill), 9% fat (Intralipid 20% Emulsion, Pharmacia Inc., Clayton, N.C.), and 81% carbohydrate (Dextrose 70% Solution, Baxter Healthcare Corp.). Essential amino acids provided one gram of body protein accretion per day, as recommended for growing swine (4, 28). The parenteral nutrition was given at half strength content for the first 24 h to assess glucose and electrolyte response, with progression to the full daily intake on day 2.

Initial ventilator settings were: fraction of inspired oxygen (FIO_2) of 0.24 to 0.30, synchronized intermittent mandatory ventilation (SIMV), breaths/minute of 10 to 12, and tidal volume of 8 ml/kg. The settings thereafter were titrated to subject response. Temperature of the inspired gas was maintained between 37 and 38°C. Suctioning of the endotracheal tube and oropharynx was done as needed. Oral care, using diluted hydrogen peroxide solution and mouth swabs, was done every four hours. Normal saline was sprayed in the snout and oral cavity to reduce mucosal drying. Subjects were repositioned from side to side every two hours to prevent pulmonary complications and skin breakdown. The ventilation pattern was altered every 20 min to one hour, with three hyperinflation breaths (150% of preset tidal volume) delivered with the ventilator or with variable, manual lung inflations, using a self-inflating resuscitation bag (Laerdal Medical, Wappingers Falls, N.Y.).

Body weight was measured every six hours, using the electronic bed scale to guide fluid and drug administration. Urine dipstick examination was done every two to four hours to monitor glucose, ketone, heme, and protein values. Passive or active-passive range of motion exercises were done for 15 min every eight to 12 h. On admission to the porcine ICU and every 12 h thereafter, catheter insertion sites were cleaned by use of povidone-iodine and were covered with transparent tape.

Cefonicid (one gram, i.v.) was given twice daily to prevent infection from indwelling catheters. Heparinized saline (100 U of porcine heparin/ml of 0.9% NaCl) was continuously infused through the carotid, femoral, and pulmonary arterial catheters at a rate of three milliliters per hour. Continuous intravenous infusion of heparin (769 U/ml) was titrated to maintain activated clotting time (ACT) between 180 and 280 sec initially. However, this degree of anticoagulation resulted in hematuria and bleeding at the intravascular insertion sites, and the target ACT value was lowered to 150 to 180 sec. Samples were measured every two to four hours in duplicate, using an automated coagulation timer (Hemochron Coagulation Analyzer, Synbiotics Corporation, San Diego, Calif.). The average value was used if the difference between results for the two samples was less than 30 sec. If the difference was 30 sec or longer, a new sample was obtained for repeat testing. Blood was drawn for hematologic and biochemical analyses on ICU days 2 and 7.

A bench top analyzer (Stat Profile Ultra, Nova Biomedical, Waltham, Mass.) was used to validate continuous arterial blood gas values. Other laboratory equipment in the porcine ICU included a centrifuge, microhematocrit centrifuge, refrigerators for storing blood products and drugs, and a blood warmer. Hematologic and blood biochemical tests and non-routine tests (e.g., culture and antimicrobial susceptibility testing of respiratory tract specimens) were done elsewhere in the institution.

A veterinary pathologist performed necropsy within a half hour to one and a half hours after euthanasia (pentobarbital sodium: ≥ 2.5 g, i.v.). Gross evaluation of the thoracic and abdominal cavities and all indwelling catheter sites was done. Electron microscopic evaluation of liver specimens taken at the time of necropsy also was done. In addition, microscopic evaluation of sections of the major thoracic and visceral organs in subject 1 was done because of concurrent disease.

Data management and analysis. Real-time physiologic data were sampled automatically during the ICU stay of each subject. Acquisition, integration, and storage of data were programmed by use of a graphic programming system (LabVIEW, National Instruments, Austin, Tex.). Digital signals from the ventilator, and intra-arterial blood gas, cardiac output, and physiologic monitors were averaged over a one-second period every second, time stamped, and stored in a spreadsheet file every one to five seconds. Continuous real-time core body temperature and $PaCO_2$ were displayed on the study computer screen to facilitate monitoring for malignant hyperthermia and appropriate sedation, respectively. Nursing ICU flow sheets were used to record fluid and drug administration, fluid output, urine dipstick examination results, body weight, interventions and subject responses, and laboratory results.

At the end of each experiment, spreadsheet files were organized, examined, and edited for each subject, using a combination of automatic and manual cleaning algorithms. Data with artifacts from thermodilution cardiac output injectate, catheter flush, bladder catheter irrigation, ventilator disconnect, and sensor change were discarded. Mean ± SD values were computed hourly for each data set.

Results

The survival outcomes are presented first to provide a framework for the physiologic, biochemical, and pathologic findings. Physiologic data are presented as mean ± SD of all hourly mean values.

Survival outcomes. Subjects 1, 2, and 3 experienced iatrogenic hemorrhagic shock from loss of arterial catheters. The catheter length, dictated by the use of the intra-arterial blood gas sensor, was inadequate to maintain catheter security, and the catheters came out within four hours of placement. Blood loss was estimated at one to two liters before emergency bedside ligation of the vessel was achieved. Hemodynamic stability was regained in subjects 1 and 2 by use of rapid infusion of one to two units of stored porcine whole blood and 0.9% normal saline.

Subject 1 survived seven days in the porcine ICU. On ICU day 2, this subject developed clinical signs of pneumonia: foul-smelling, yellow, purulent, copious secretions from the snout, oropharynx, and endotracheal tube; fever; and hypoxemia. Culture of respiratory tract aspirates yielded *Bordetella bronchiseptica* and *Pasteurella multocida*. Despite additional antibiotic coverage using ceftazidime, vancomycin, and ciprofloxacin, this subject developed intractable hypoxemia. Large minute volumes, positive end-expiratory pressure, 100% oxygen, hourly position changes (including prone positioning), chest physiotherapy, and furosemide administration were needed to maintain oxygenation. When the subject failed to respond to treatment, it was euthanized by administration of pentobarbital (five grams, i.v.).

Subject 2 survived five and a half days in the porcine ICU. During weaning from mechanical ventilation, this pig experienced sudden cardiovascular collapse with electromechanical dissociation. Blood pressure decreased precipitously, from 118/80 mmHg to 22/7 mmHg, with atrial-ventricular dissociation in the V leads; pupils were fixed and dilated.

Subjects 3 and 4 survived 44 and 41 h, respectively, in the porcine ICU until euthanasia at the planned termination of the experiment. Porcine blood was unavailable to treat hemorrhagic shock in subject 3, and crystalloid (0.9% saline) and synthetic colloid (Hextend, Abbott Laboratories, Chicago, Ill.) were infused rapidly. Despite frequently administered normal saline bolus infusions, the subject remained hypotensive and alternatively had

Table 1. Physiologic parameters in domestic farm pigs (n = 4) while they were in the experimental porcine ICU

Physiologic parameter	Subject 1	Subject 2	Subject 3	Subject 4
Heart rate (beats/min)	95 ± 3	111 ± 3	110 ± 6	97 ± 6
Mean arterial pressure (mmHg)	75 ± 4	90 ± 4	57 ± 3.9	117 ± 6.4
Central venous pressure (mmHg)	8.9 ± 1.2	7 ± 1	1 ± 0.53	0 ± 0.54
Arterial temperature (°C)	40.4 ± 0.1	40.6 ± 0.07	38.3 ± 0.34	39.5 ± 0.15
Peripheral oxygen saturation (%)	93.5 ± 2	95.7 ± 1	94.6 ± 2.6	96.2 ± 3.2
pH, arterial	7.42 ± 0.02	7.48 ± 0.01	7.41 ± 0.01	7.39 ± 0.01
Carbon dioxide, arterial (mmHg)	48.5 ± 1.7	42.9 ± 1.1	51 ± 0.97	49 ± 1.7
Oxygen, arterial (mmHg)	91 ± 7.8	86 ± 3.9	120 ± 10.2	126 ± 17
Respiratory rate (mandatory and spontaneous [breaths/min])	18 ± 2	18 ± 2	11 ± 1	11 ± 1
Minute volume, total (L/min)	14.9 ± 0.85	12.2 ± 1.3	5.7 ± 0.28	6.6 ± 0.30
Tidal volume (ml)	829 ± 0.06	684 ± 0.07	530 ± 0.03	638 ± 0.03
Peak inspiratory pressure (cmH_2O)	35 ± 2	20 ± 2	19 ± 1.4	5 ± 1.2

Data are expressed as mean ± SD of all 60-min averages.

Table 2. Selected hematologic and biochemical values of porcine subjects (n = 4) and normal ranges for a 70-to 100-kg pig (16)

Subject	1		2		3			4	
Parameter (Normal range)	ICUd2	ICUd7	ICUd2	Preopd10	OR	ICUd2	Preopd10	ICUd1	ICUd2
WBC ($\times 10^3/\mu l$) (10.3-20.7)	21.9*	11.1	28.7*	20.4	18	17.9	14.1	16.9	18.6
Hemoglobin (g/dl) (12.5-13.5)	7.8*	4.8*	10.9*	12.9	10.6*	5.3*	10.9*	10.5*	11.5*
Hematocrit (%) (42-44)	21.9*	14.3*	30.8*	39.4*	32.7*	16.9*	34.4*	31.8*	35.4*
Platelets ($\times 10^3/\mu l$) (232-368)	109*	138*	203	277	293	181*	260	187*	233
Creatinine (mg/dl) (1.0-2.6)	2.4	2.2	2.2		1.7	2.2		4.2*	2.9*
Glucose (mg/dl) (60-136)	73	99	100		NA	NA		NA	NA
Sodium (meq/L) (135-152)	140	135	140		139	141		137	137
Chloride (meq/L) (94-106)	102	89*	102		96	108*		100	99
Potassium (meq/L) (4.9-7.1)	NA	4.1*	NA		3.7*	4.1*		4.2*	3.9*

*Abnormal value; ICUd = ICU day; NA = not available; OR = operating room; Preopd = preoperative day ; Blanks = chemistries not done preoperatively.

episodes of bradycardia and supraventricular tachycardia. The pig never regained consciousness after surgery, and reflexes were not elicited in response to stimulation. Subject 4 did not experience complications.

Cardiopulmonary and biochemical responses. The physiologic parameters for each subject are shown in Table 1. The heart rate was similar across subjects, and averaged 95 to 111 beats per minute. Mean arterial pressure varied widely, and was considerably lower in subject 3 who was hypotensive throughout the experiment. Core body temperature, measured in a central artery, was higher in subjects 1 and 2. Likewise, total respiratory rate (mandatory and spontaneous) and minute volume were higher in the first two subjects. Tidal volume was higher in subject 1 due to the use of large tidal volumes to treat progressive hypoxemia and hypercapnea; peak inspiratory pressure consequently was highest in this subject.

Preoperative, average daily ICU, and postmortem weight changes varied from one to four kilograms. Weight was stable during the full and abbreviated protocols, and no subject lost weight in the ICU. Average hourly intravenous intake from all sources (e.g., catheter flush solutions, parenteral nutrition, crystalloids, resuscitation fluids) and urine output were as follows: 167 and 87 ml/h (subject 1), 179 and 112 ml/h (subject 2), 299 and 240 ml/h (subject 3), and 180 and 94 ml/h (subject 4). Net positive fluid balance was 11,970, 8,468, 575, and 3,342 ml, respectively, in subjects 1–4.

Table 2 displays selected hematologic and biochemical values for each subject. Subjects 1 and 2 had high white blood cell count in the ICU, except day 7 when subject 1 had been given additional antibiotic therapy to which the gram-negative organisms were sensitive. Because of postmortem evidence of pneumonia in the first two subjects, we added to the protocol for subsequent subjects a preoperative complete blood count with manual differential count to diagnose pre-existing infection. In the ICU, hemoglobin and hematocrit values in subjects 1 and 3 were markedly decreased. Creatinine concentration was high in subject 4. Potassium values were low in all subjects, despite supplemental potassium in the parenteral nutrition or crystalloid infusion.

Figure 1 shows the daily sedation requirements for each subject. Daily requirements increased in every case, except in subject 3, who was comatose. Figure 2 displays the average hourly amount of heparin infused each day and ACT ranges for each subject. As evidenced by the narrower range of ACTs, stability in anticoagulation was achieved between the third and fourth ICU days for the full protocol. Subject 1 required an average heparin dose of 89 U/kg/h, and higher heparin requirements were associated with increasing morbidity. In contrast, heparin requirements were more stable in subject 2; average heparin dose was 76 U/kg/h. The average heparin dose in subjects 3 and 4 was 47 and 43 U/kg/h, respectively.

Gross necropsy and microscopic findings. Notable abnormal findings on necropsy are presented for each subject in Table 3. Microscopic findings in subject 1 were as follows. Small number of lymphocytes and plasma cells were observed ran-

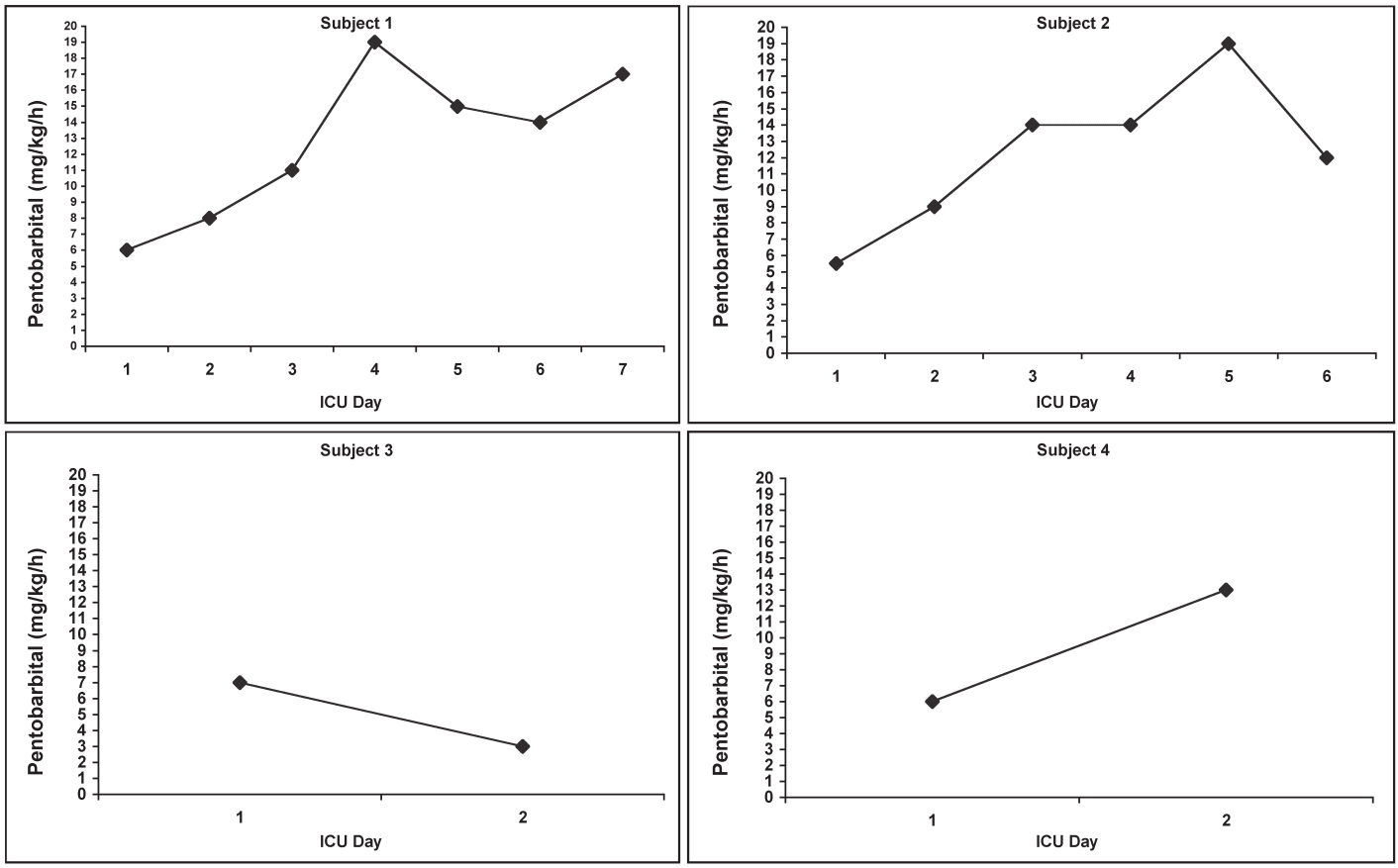

Figure 1. Daily pentobarbital requirements (mg/kg/h) of each subject in the porcine intensive care unit (ICU; n = 4). Notice that day 6 represents 12 h for subject 2, and day 2 consists of 20 h for subject 3 and 17 h for subject 4.

domly within the interstitium of the kidney. There was mild, diffuse congestion of the spleen. Rare to a few foci of wavy fiber change were evident within the midmural myocardium; these fiber changes appeared frequently in the sections of interventricular septum and right ventricular free wall. As well, patchy areas of myocyte necrosis were observed within the inner third of the myocardium. The lungs had marked expansion of alveolar septa, with resultant collapse of alveoli. In all sections of lung, there was severe consolidation; hyaline membrane formation; fibrosis; infiltrating macrophages, neutrophils, and lymphocytes; type-II pneumocyte hyperplasia; cellular debris; a few foci of gram-negative bacteria; and fibrin thrombi.

Before electron microscopic evaluation, the liver specimen for subject 2 was lost during severe flooding of the Texas Medical Center; thus, electron microscopic findings are reported for only three subjects. Ultrastructural findings of the liver in subject 1 consisted of irreversible organelle changes: chromatin dispersion in nuclei, and focal calcium precipitation in mitochondria. Subjects 3 and 4 had mild, reversible organelle changes indicating membrane damage (i.e., vacuoles with membrane whorls) and effaced mitochondria cristae (mitochondriosis), which are potentially reversible alterations.

Discussion

The purpose of these experiments was to test the feasibility of maintaining survival of healthy but mechanically ventilated swine for longer than three days under clinical ICU conditions. The experiments were only partially successful. We were unable to keep the subjects healthy, but we were able to maintain survival of critically ill swine for longer than three days. Our porcine ICU model was an excellent representation of the clinical adult ICU. Although we used growing-finishing swine, body weight was equivalent to that of the average adult human and we could replicate, to a large extent, clinical care protocols. As is the case in the clinical setting, the interactive effects of treatment, preexisting and concomitant disease, and the ICU environment complicated care of the subjects. Thus, our model may prove beneficial for short-term study of iatrogenesis.

We documented that it may be feasible to maintain survival of mechanically ventilated swine in the ICU for longer than three days; however, only one subject advanced to ventilator weaning, and it died before weaning was completed. Before it can be used for studies on weaning, further research with the model is needed to reproduce and extend survival time and reduce iatrogenic complications. Despite hemorrhagic shock requiring bedside surgery and resuscitation, and postmortem evidence of pneumonia in two subjects, our model generated hemodynamic, arterial blood gas, and respiratory values similar to those seen in ICU patients with these conditions and undergoing these therapies.

Belanger and colleagues (3) recently reported successful cardiopulmonary bypass in 80-kg pigs for up to 22 h; however, use of a large-animal ICU model that closely mimics the clinical ICU has not been reported to our knowledge. There are many

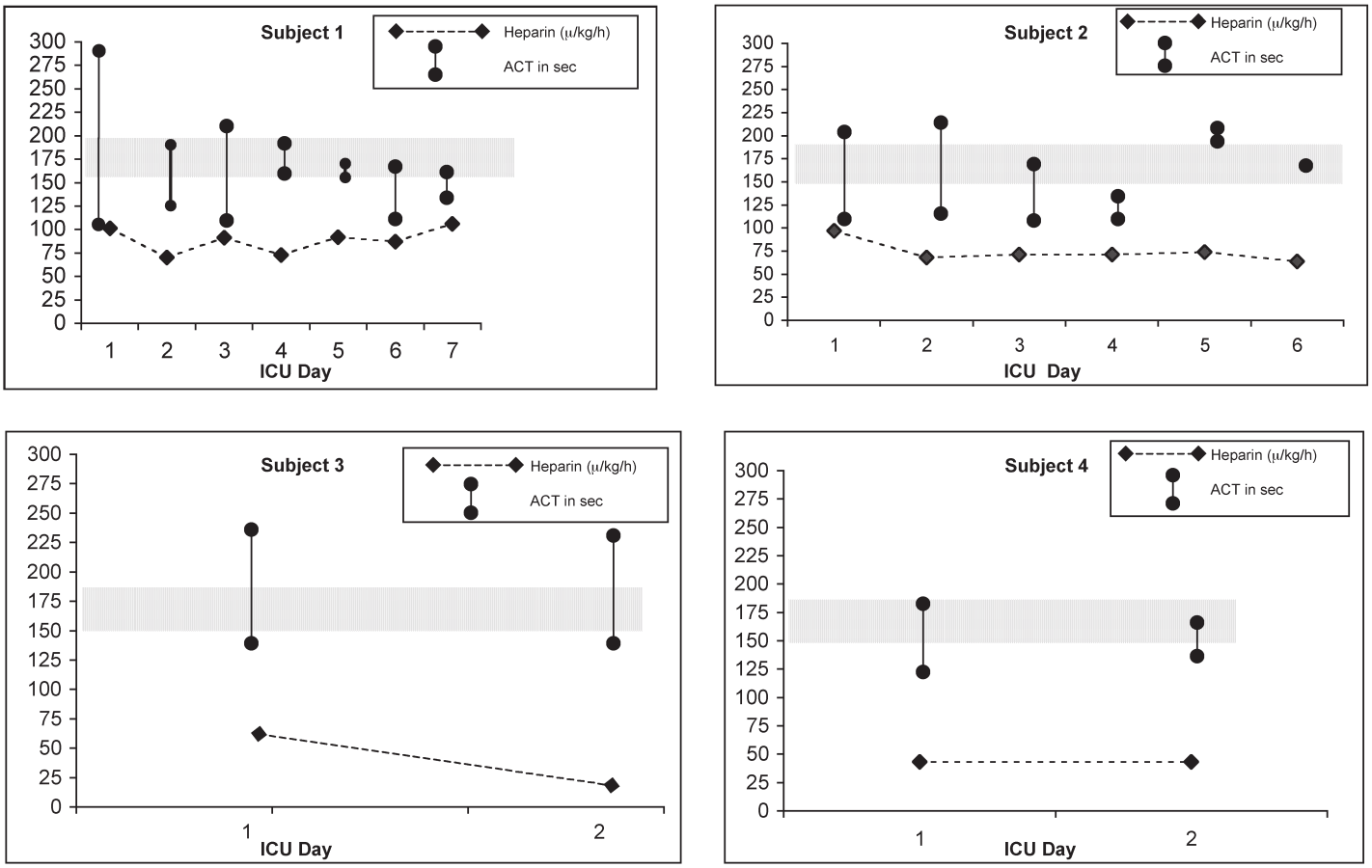

Figure 2. Average amount of heparin infused (U/kg/h) and range of activated clotting time (ACT) in seconds for each subject by ICU day (n = 4). The shaded area represents the target range (150 to 180 seconds) for ACT.

advantages to being able to study pigs weighing ≥ 70 kg in the ICU. Pigs have cardiovascular, skin, musculoskeletal, and hepatic anatomic similarities to those of humans. Likewise, there are similarities in porcine and human hemodynamics; response to shock; wound healing; and renal, hematologic, and metabolic physiology (11, 18, 25). Thus, a porcine adult ICU model would be valuable in the study of conditions commonly seen in critically ill patients. A novel feature of our model is that the pigs were in bed. Complete bed rest adds the important horizontal dimension of immobility that is constant in critically ill patients, and one that is not seen in long-term ventilated large animal models (1).

The stresses observed using this protocol were similar to those of a canine ICU model (7), and morbidity attributable to pneumonia, pulmonary microemboli, and hypotension was similar. Compared with the canine model, our porcine model had marginally less barotrauma (one pig [25%] versus five dogs [29%]), and in both models, the barotrauma was ventilator induced. We used a lower tidal volume (8 ml/kg versus 15 ml/kg) to avoid barotrauma, and altered ventilation to minimize non-compression atelectasis. Nevertheless, postmortem findings of pneumonia despite absence of clinical signs of disease in subject 2 suggests that, with longer ICU stays, pneumonia and subsequent barotrauma are likely to be common problems with use of the porcine adult ICU model.

Although the lungs were unremarkable in subjects 3 and 4, the ICU stay was less than 72 h, the time when nosocomial pneumonia becomes evident in critically ill patients. It is not clear whether the pigs from the open herd had sub-clinical pneumonia at the time of transport or developed it in the research facility. Because all of our pigs appeared healthy, it may be beneficial to evaluate white blood cell count and/or cytokine values before committing the animal to the ICU model. The cost of such a preoperative evaluation would be far less than preparing the animal for ICU experimentation and might elucidate the source of pneumonia.

Although sepsis was a problem in the canine model (7), the major problem in this feasibility study was hemorrhagic shock from loss of arterial catheters. We view continuous intra-arterial blood gas monitoring as essential for ICU management. Aside from limiting the amount of blood withdrawn in a protocol requiring frequent sampling to determine blood gas tensions, real-time $PaCO_2$ was enormously helpful in titrating sedation and managing ventilator settings. Likewise, real-time pHa and the calculated values of bicarbonate and base excess were useful in guiding the treatment of hemorrhagic shock. The constraint on the length of the arterial catheter in the first two subjects resulted in a mere one centimeter of catheter tip in situ; this placement was clearly inadequate. We attribute the arterial catheter dislodgment in subject 3 to the friability of porcine vessels and a new surgical instrumentation team.

Resuscitation from hemorrhagic shock was successful in the subjects that had porcine blood transfusions. The pig that was resuscitated by use of crystalloid and colloid remained hypotensive and had a critically low hemoglobin value. Although subject

Table 3. Necropsy findings in domestic farm pigs (n = 4) after 41 to 168 h in the experimental porcine ICU

Subject	Time in ICU	Gross findings
1	168 h (7 d)	• Mucosal erosions (1 × 3 mm) in laryngeal area and (1 × 0.5-cm) at first tracheal cartilage ring • Yellowish exudate in primary bronchi and trachea • Slight enlargement of tracheobronchial lymph nodes • Mediastinal emphysema • Heavy, wet, dark red appearance of both lungs • Diffuse consolidation of all lung lobes; small regions of dorsal left and dorsal right cranial lobes aerated • Dark amber free fluid (600 ml) in abdominal cavity • Multiple fecoliths in distal portion of colon • Multiple ecchymotic hemorrhages in bladder mucosa • Small fibrin tags adhered to pulmonary artery catheter • Thrombus at femoral venous and arterial catheter tips
2	132 h (5.5 d)	• Ulcer (6 × 3 mm) in epiglottis • Bilateral ulceration and pseudomembrane formation before first tracheal ring • Diffuse lung consolidation, edema, and interstitial pneumonia • Clear fluid (50 ml) in pericardial sac • Fibrin tags at hilus • Acute thrombus, left contributory pulmonary artery • Serosanguinous fluid (300 ml) in peritoneal cavity • Localized pinpoint hemorrhagic areas in bladder wall • Thrombi at all catheter insertion sites
3	44 h (1.8 d)	• Focal area of ulcerations on trachea • Serosanguinous fluid (100 ml) in abdominal cavity • Multiple petechiae on mucosal surface of bladder
4	41 h (1.7 d)	• Focal ulceration of bladder mucosa

d = day; ICU = intensive care unit.

1 also had critically low hemoglobin concentration, it was hemodynamically stable after resuscitation on ICU day 1 until ICU day 7 when hypoxemia and acidosis became overwhelming. Controversy over the preferred agent for volume resuscitation has yet to be resolved. Animal models of hemorrhagic shock have had equivalent or improved survival in response to administration of lactated Ringer's, compared with normal saline solution (12); however, some authors have suggested that the hypotonicity of Ringer's solution may be a concern with respect to interstitial edema (23). Hextend prolonged survival in a rat model with septic shock (14) when compared with saline and lactated Ringer's solutions. Short-term improvements in hemodynamic function have been observed after resuscitation with hypertonic solutions; however, the improvements were not sustained (27).

Replacement volume affects resuscitation outcome. Traverso and colleagues (26) reported progressive improvements in porcine survival, using 100 and 300% replacements of blood loss with normal saline. Subject 3 of the study reported here had 200% replacement, which was sufficient to maintain survival but not hemodynamic stability. Most resuscitation studies in animal literature were short term (< 12 h), and our experience with the porcine ICU model suggests that porcine blood is the preferred agent for use in resuscitation from hemorrhagic shock.

On postmortem examination, subject 1 had a gastric ulcer. Stress-related mucosal damage of the gastrointestinal tract is common in the critically ill patient (24), and gastric and duodenal ulcers have been reported in piglets following hemorrhage (10, 15, 22). Three subjects did not have gross abnormalities in gastric mucosa. Subject 1 was the only pig with a sustained orogastric tube in situ, and the tube may have contributed to the focal gastric erosion. We did not administer gastric acid suppression agents or enteral feedings, two commonly used protective strategies in animals (7) and humans (24) on mechanical ventilation.

We chose parenteral instead of enteral nutrition for several reasons. Continuous sedation would impair gastric motility and, consequently, meeting of the nutritional goals. Absent bowel sounds and the presence of fecoliths in the bowel on postmortem examination confirmed impaired gastrointestinal motility. Nasogastric tubes theoretically stent the esophageal sphincter, predisposing the pig to aspiration, and confirmed placement of a postpyloric feeding tube was beyond the resources available to this feasibility study. Most critically ill patients experience gastrointestinal tract complications—high gastric residuals, constipation, diarrhea, abdominal distention, vomiting, and regurgitation—in association with gastric enteral feedings (19). We wanted to avoid such complications and their attendant risk for aspiration pneumonia.

Postmortem examination did not reveal any evidence of fat atrophy, indicating adequate nutrition of our subjects. Postmortem body weight was greater than preoperative body weight for every subject, with the weight gain being greater (approx. four kilograms) in subjects 1 and 2. However, we caution equating weight gain with adequate nutrition because we don't know how much of the gain was because of volume overload and/or fluid in the extravascular spaces. Net fluid balances were high for subjects 1 and 2, but good urine output was maintained in all subjects, including subject 3 with clinical signs of hypovolemia and marginal net fluid balance.

In conclusion, results of this feasibility study documented that swine weighing 70 kg or more can survive mechanical ventilation in an ICU for longer than three days. Furthermore, survival from complications of iatrogenic hemorrhage, stress-related mucosal injury, hypokalemia, and acute respiratory failure is feasible by using clinical care protocols. Therefore, this porcine intensive care model may be useful for exploring the safety and effectiveness of critical care interventions in the animal laboratory before clinical trials are done. Further experience with the porcine adult ICU model is needed to develop a reproducible model for healthy and critically ill swine, extend the survival time, and reduce iatrogenic complications.

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References

1. **Alpard, S. K., J. B. Zwischenberger, W. Tao, D. J. Deyo, D. L. Traber, and A. Bidani.** 2000. New clinically relevant sheep model of severe respiratory failure secondary to combined smoke inhalation/cutaneous flame burn injury. Crit. Care Med. **28:**1469-1476.
2. **Becker, B. A., J. J. Ford, R. K. Christenson, R. C. Manak, G. L. Hahn, and J. A. DeShazer.** 1985. Cortisol response of gilts in tether stalls. J. Anim. Sci. **60(1):**264-270.
3. **Belanger, M., C. Wittnich, S. Torrance, and S. Juhasz.** 2002. Model of normothermic long-term cardiopulmonary bypass in swine weighing more than eighty kilograms. Comp. Med. **52(2):**117-121.
4. **Fuller, M. F., R. McWilliam, T. C. Wang, and L. R. Giles.** 1989. The optimum dietary amino acid pattern for growing pigs. 2. Requirements for maintenance and for tissue protein accretion. Br. J. Nutr. **62(2):**255-267.
5. **Griffith, M. K. and J. E. Minton.** 1992. Effect of light intensity on circadian profiles of melatonin, prolactin, ACTH, and cortisol in pigs. J. Anim. Sci. **70:**492-498.
6. **Gronert, G. A., D. L. Fung, S. C. Haskins, and E. P. Steffey.** 1999. Deep sedation and mechanical ventilation without paralysis for 3 weeks in normal beagles. Anesthesiology. **90(6):**1741-1745.
7. **Gronert, G. A., S. C. Haskins, E. P. Steffey, and D. Fung.** 1998. Plasma electrolyte and metabolite concentrations associated with pentobarbital or pentobarbital-propofol anesthesia during three weeks' mechanical ventilation and intensive care in dogs. Lab. Anim. Sci. **48(5):**513-519.
8. **Hahn, G. L.** 1989. Body temperature rhythms in farm animals: a review and reassessment relative to environmental influences. Proceedings of the 11th ISB-Congress. 325-337. The Hague: SPB Academic Publishing.
9. **Hanneman, S. K. G., G. L. Ingersoll, A. R. Knebel, M. E. Shekleton, S. Burns, and J. M. Clochesy.** 1994. Weaning from short-term mechanical ventilation: a review. Am. J. Crit. Care. **3:**421-443.
10. **Hannon, J. P.** 1992. Hemorrhage and hemorrhagic shock in swine: A review, p. 197-245. *In* M. M. Swindle (ed.), Swine as models in biomedical research. Iowa State University Press, Ames, Iowa.
11. **Hart, B. B., G. G. Stanford, M. G. Ziegler, C. R. Lake, and B. Chernow.** 1989. Catecholamines: study of interspecies variation. Crit. Care Med. **17:**1203-1222.
12. **Healey, M. A., R. E. Davis, F. C. Liu, W. H. Loomis, and D. B. Hoyt.** 1998. Lactated Ringer's is superior to normal saline in a model of massive hemorrhage and resuscitation. J. Trauma **45(5):**894-899.
13. **Holliman, C. J., K. Kenfield, E. Nutter, J. R. Saffle, and G. D. Warden.** 1982. Technique for acute suprapubic catheterization of the urinary bladder in the pig. Am. J. Vet. Res. **43:**1056-57.
14. **Kellum, J. A.** 2002. Fluid resuscitation and hyhperchloremic acidosis in experimental sepsis: improved short-term survival and acid-base balance with Hextend compared with saline. Crit. Care. Med. **30(2):**300-305.
15. **Kivilaakso, E., J. Ahonen, K-F. Aronsen, K. Hockerstedt, T. Kalima, M. Lempinen, H. Suoranta, and E. Vernerson.** 1982. Gastric blood flow, tissue gas tension and microvascular changes during hemorrhage-induced stress ulceration in the pig. Am. J. Surg. **143(3):**322-330.
16. **Knebel, A. R., M. E. Shekleton, S. Burns, J. M. Clochesy, S. K. G. Hanneman, and G. L. Ingersoll**. 1994. Weaning from short-term mechanical ventilation: concept development. Am J. Crit. Care. **3:**416-420.
17. **Minton, J. E., D. L. Davis, and J. S. Stevenson.** 1989. Contribution of the photoperiod to circadian variations in serum cortisol and melatonin in boars. Domest. Anim. Endocrinol. **6(2):**177-181.
18. **Mitruka, B. M., and H. M. Rawnsley.** 1981. Clinical biochemical and hematological reference values in normal experimental animals and normal humans, 2nd ed. Masson Publishing, Inc., New York.
19. **Montejo, J. C.** 1999. Enteral nutrition-related gastrointestinal complications in critically ill patients: a multicenter study. The Nutritional and Metabolic Working Group of the Spanish Society of Intensive Care Medicine and Coronary Units. Crit. Care Med. **27(8):**1447-1453.
20. **National Institutes of Health.** 1985. Principles of laboratory animal care (NIH publication No. 86-23, revised version). National Institutes of Health, Bethesda, Md.
21. **National Research Council.** 1996. Guide for the care and use of laboratory animals. National Academy Press, Washington, D.C.
22. **Norton, L., P. Nolan, J. E. L. Sales, and B. Eiseman.** 1972. A swine stress ulcer model. Ann. Surg. **176(2):**133-138.
23. **Prough, D. S. and A. Bidani.** 1999. Hyperchloremic metabolic acidosis is a predictable consequence of intraoperative infusion of 0.9% saline. Anesthesiology **90(5):**1247-1249.
24. **Steinberg, K. P.** 2002. Stress-related mucosal disease in the critically ill patient: risk factors and strategies to prevent stress-related bleeding in the intensive care unit. Crit. Care Med. **30(6 suppl):**S362-364.
25. **Swindle, M. M.** 1985. Porcine models in surgical research: an overview, p. 235-242. *In* M. E. Tumbleston (ed.), Swine in biomedical research. Plenum Publishing, New York.
26. **Traverso, W. L., W. P. Lee, and M. J. Langford.** 1986. Fluid resuscitation after an otherwise fatal hemorrhage: I. Crystalloid solutions. J. Trauma **26(2):**168-175.
27. **Wade, C. E., J. P. Hannon, C. A. Bossone, M. M. Hunt, J. A. Loveday, R. Coppes, and V. L. Gildengorin.** 1989. Resuscitation of conscious pigs following hemorrhage: comparative efficacy of small-volume resuscitation. Circ. Shock **29:**193-204.
28. **Wang, T. C. and M. F. Fuller.** 1989. The optimum dietary amino acid pattern for growing pigs. 1. Experiments by amino acid deletion. Br. J. Nutr. **62(1):**77-89.