Estimating Mortality Risk in Preoperative Patients Using Immunologic, Nutritional, and Acute-Phase Response Variables

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We measured the delayed type hypersensitivity (DTH) skin test response, along with additional variables of host immunocompetence in 245 preoperative patients to determine which variables are associated with septic-related deaths following operation. Of the 14 deaths (5.7%), 12 were related to sepsis and in 2 sepsis was contributory. The DTH response ($p < 0.00001$), age ($p < 0.0002$), serum albumin ($p < 0.003$), hemoglobin ($p < 0.02$), and total hemolytic complement ($p < 0.03$), were significantly different between those who died and those who lived. By logistic regression analysis, only the DTH skin test response (log likelihood = 41.7, improvement $X^2 = 6.24$, $p < 0.012$) and the serum albumin (log likelihood = 44.8, improvement $X^2 = 17.7$, $p < 0.001$) were significantly and independently associated with the deaths. The resultant probability of mortality calculation equation was tested in a separate validation group of 519 patients (mortality = 5%) and yielded a good predictive capability as assessed by (1) $X^2 = 0.08$ between observed and expected deaths, NS; (2) Goodman-Kruskall G statistic = 0.673) Receiver-Operating-Characteristic (ROC) curve analysis with an area under the ROC curve, $A_z = 0.79 \pm 0.05$. We conclude that a reduced immune response (DTH skin test anergy) plus a nutritional deficit and/or acute-phase response change are both associated with increased septic-related deaths in elective surgical patients.

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THE ABILITY TO IDENTIFY PATIENTS who are at increased risk for postoperative complications of sepsis and death is advantageous, particularly if steps could be taken to reduce this risk. Several approaches to this problem have been taken, both for the perioperative risk period, encompassing the first 24 hours after surgery, and the postoperative period, encompassing the next 30 days after surgery. Thus far assessment of risk during the second period has focused on the relation between nutritional deficits and clinical outcome. Although it is accepted that postoperative sepsis contributes significantly to an adverse outcome, risk assessment analysis that takes into consideration the adequacy of antibacterial host defense mechanisms is lacking.

The association between abnormal host defense as reflected by the delayed type hypersensitivity skin test response to common antigens and an unfavorable outcome after hospitalization was initially shown by MacLean et al.[1] and later elaborated by Meakins et al.[2] Additional laboratories have confirmed the hypothesis that a reduced delayed type hypersensitivity skin test response on hospital admission identifies surgical patients with increased chances of mortality, usually secondary to sepsis.[3-14] One group of investigators does not concur with this hypothesis.[15] Still others believe that clinical judgment is better than measurements of the delayed type hypersensitivity skin test response as a means of identifying those patients at increased risk for septic-related death.[16]

This controversy is attributed to three problems. First, the technique of delayed type hypersensitivity skin testing. The type and number of antigens used are not uniform.[17] Second, most of these studies fail to distinguish between preoperative elective patients and those seen after major trauma, sepsis, or burns. Third, most of these studies have considered a single variable, delayed type hypersensitivity skin test response, without controlling for possible confounding variables such as patient age, nutritional status, acute phase response, and various aspects of specific and nonspecific host defense function.

For these reasons, measurement of specific and nonspecific host defense functions, along with measures of the serum albumin and circulating C-reactive protein, were carried out in elective preoperative patients admitted

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for gastrointestinal surgery, in order to determine those variables that significantly contribute to the estimate of risk for septic-related death in the postoperative period. The delayed type hypersensitivity skin test response and the serum albumin were the only variables that significantly and independently contributed to the estimate of this risk. Given these variables in a preoperative patient, a probability of septic-related death can be calculated, which can assist the surgeon in the clinical management of the patient.

Methods

Subjects

Patients admitted for elective resections of the esophagus, stomach, small intestine, large intestine, or rectum were entered into the study. This study was approved by the Ethics Committee on Human Experimentation of the Royal Victoria Hospital, McGill University. Exclusion criteria were chronic or acute steroid administration, recent transfusion with blood or blood products, chemotherapy, radiotherapy, and/or emergency surgery.

Variables Measured

The following variables were measured on admission to hospital and before surgery. The Acute Physiology And Chronic Health Evaluation (APACHE II) score was calculated according to the method of Knauss et al.[18] Circulating blood lymphocyte, monocyte, and polymorphonuclear neutrophil counts (in cells$\times 10^9$/L) and the blood hemoglobin concentration (g/L) were carried out in the clinical laboratory. Serum albumin (g/L), circulating total immunoglobulin (g/L), IgA (g/L), IgG (g/L), IgM (g/L) were measured by high-voltage paper electrophoresis. Serum C-reactive protein, complement C3 (g/L), and total hemolytic complement CH50 (%) were measured by nephelometry.

Leukocyte Adherence

Leukocyte adherence to nylon wool was measured using a modification of the technique of McGreggor et al.[19] as published elsewhere.[20] Briefly, 1 ml of blood kept at 37 C was passed under gravity through a 1-ml tuberculin syringe fitted with a 26-gauge, 5/8 inch needle and packed with 30 mg of nylon fiber (Leuko-Pack Leukocyte Filter, Fenwal Laboratories, Deerfield, IL) pre-wetted with 1 ml of minimal essential medium (Gibco) containing 10 mM HEPES buffer at pH 7.4 kept at 37 C. White cell counts of the starting blood and the effluent blood were performed with an automated cell counter (Coulter Electronics Inc., Model Z_f, verified by manual counts) and adherence of cells was calculated and expressed as a percentage of white cells adhering to the filter.

Polymorphonuclear Neutrophil Chemotaxis

Polymorphonuclear neutrophil chemotaxis was measured using zymosan activated serum as the chemoattractant and a modification of the 48-well nucleopore technique of Falk et al.[21] Polymorphonuclear neutrophils were obtained using ficoll-hypaque sedimentation of heparinized whole blood. Red blood cells contaminating the polymorphonuclear neutrophil preparation were lysed with 0.9% amonnium chloride. Cells were resuspended to a concentration of 10^6/ml in minimal essential medium containing 10 mM HEPES buffer at pH 7.4 prewarmed to 37 C, and 0.1 ml was added to the top wells of a 48-well assembly that was separated into upper and lower chambers by a 10- micron thick filter (Nucleopore Corporation) with 3-micron pore size. The bottom wells contained prewarmed pooled zymosan activated serum prepared from healthy subjects. The same pool of ZAS was used for the duration of the study, frozen in aliquots at −70 C. There was no loss of chemoattraction potential during the course of the study period as verified with control neutrophils checks. The chambers were incubated at 37 C for 30 minutes, the filters were removed, fixed, and stained with hematoxyline and eosin. The cells reaching the opposite side of the filter were counted and chemotaxis was expressed as the number of cells per high-power field that had migrated to the opposite side of the filter.

Delayed Type Hypersensitivity Skin Tests

Skin tests were performed on the outer aspect of the upper arm or volar surface of the forearm. The five common antigens used were Candida (Candidin 1:100 dilution), Mumps skin test antigen (undiluted), purified protein derivative (5 TU), Trichophytin, and Streptokinase-Streptodornase (Varidase, 100 units/ml). They were injected intradermally in a volume of 0.1 ml. A 0.1-ml saline control was also used. The two greatest diameters of the resulting induration for each antigen at 24 or 48 hours were measured, summed, divided by two, and the resulting mean was recorded. The erythema surrounding the induration was ignored. The results were expressed as a DTH score for each patient, which is defined as the sum of the mean diameters of induration of each individual antigen response, minus the saline control, as measured above. Alternately, an antigen response was defined as positive if the mean diameter of the induration for that antigen was equal to or greater then 5 mm. Subjects were then classified as reactive if they had two or more positive responses, were relatively anergic with one antigen re-

sponse, and were anergic if they demonstrated no response to any antigen.

Preoperative, Perioperative, and Postoperative Follow-up

Subject age, the admitting diagnosis, type and duration of surgery, perioperative blood, and blood-product administration were recorded. Patient follow-up extended to 30 days after surgery. Major sepsis was defined as a proved intracavitary abscess, bacteremia, ascending cholangitis, or fatal pneumonia. Autopsies were requested from the next of kin for all patients who died and this was granted in 10 of 14 cases (71.4%). The cause of death was thus determined from the autopsy report or the death certificate of the patient when autopsy was refused. Deaths were classified as septic-related if the cause of death was due to overwhelming sepsis, or if the patient died following a major sepsis episode. Otherwise the death was classified as nonseptic. Urinary tract infections, infections of wounds and soft tissues, and pneumonias that did not contribute to death (based on autopsy) were classified as minor infectious complications.

Statistical Analysis

All statistical analysis were carried out using the BMDP-PC software.[22] Data are shown as mean ± sd. Analysis of variance was used to compare means of continuous variables between the reactive, nonreactive, and control subjects. Significant differences between any two groups was tested by multiple t-tests with Bonferroni corrections. The unpaired t-test was used to compare continuous data between patients who survived and those who died. Significant differences of categorical deaths among patient groups was determined by Chi-square analysis with Yates' correction where appropriate. Stepwise logistic regression analysis was used to examine which variables were significantly associated with hospital deaths, and to estimate the likelihood that a patient would die if he or she were operated on, given these variables. The logistic function takes the following form:

$$\Pr[\text{outcome} \mid x_1, x_2, \ldots, x_k] = 1/\{1 + e^{(-b_0 - b_1 x_1 - \cdots - b_k x_k)}\} \quad (1)$$

where: Pr = probability of a particular outcome given independent variables $x_1, x_2, \ldots, x_k$, e = the exponential function (natural logarithm), b_0, $b_1 \cdots b_k$ are the coefficients to be estimated, and $x_1, \cdots x_k$ are the independent variables.

Starting with the constant model (i.e., b_0 as the only parameter), a variable was entered into the equation only if its addition increased the likelihood of the model with a significant p value (inclusion and exclusion limits were set at $p < 0.05$) using two-tailed tests of significance. This analysis was carried out using a training set of 245 patients collected between July 1, 1986 and December 31, 1987. Incidence of mortality was used as the dependent variable. The independent variables were (1) patient age; (2) delayed type hypersensitivity skin test score; (3) APACHE II score minus the age points; (4) sex entered as man = 1, woman = 2; (5) leukocyte adherence; (6) polymorphonuclear neutrophil chemotaxis; (7) circulating polymorphonuclear neutrophil count; (8) circulating lymphocyte count; (9) circulating monocyte count; (10) total serum immunoglobulin; (11) serum IgG; (12) serum IgM; (13) serum IgA; (14) C-reactive protein; (15) circulating serum albumin; (16) serum C3 levels; (17) serum CH50 levels; and (18) circulating hemoglobin level.

A modification of the "prospective independent study" method was used in order to test for the precision of the resultant model to predict a hospital death.[23] A separate validation data set of 519 patients matched for age, diagnosis, and type of surgery was procured from a retrospective analysis of prospectively collected data between January 1, 1980 to December 31, 1984. This patient data base forms part of our continuing studies of host defense abnormalities of surgical patients begun in 1977 at our institution. A probability of death was calculated for each patient using the logistic regression equation derived from the training set. The patients were then divided into deciles of risk based on this probability value. The number of expected deaths within each decile was obtained by summing the individual probabilities. This expected mortality rate was compared to the actual deaths, and differences were tested for significance by Chi-square analysis. The Goodman-Kruskel G statistic was also calculated in order to measure the predictive power of this index. Analogous to a correlation coefficient, its values range from −1 to +1, with values near +1 indicating a strong positive correlation, −1 indicating a strong negative correlation, and a value of 0 indicating no correlation.[24] The performance of this probability of death equation in predicting outcome was assessed by Receiver-Operating Characteristic (ROC) curve analysis. The ROC curve demonstrates the relation between those correctly predicted to be nonsurvivors (true positives) and the percentage of those incorrectly predicted to be survivors (false positives) as the criterion for each status is systematically varied. The area beneath the ROC curve, A_z, is used as an index of predictor performance. Az ranges from 0.5 for chance performance to 1 for perfect prediction.[25]

Results

The demographic data and types of surgery for the training set of 245 patients used in the analysis are sum-

TABLE 1. *Demographic Characteristics and Type of Surgery of the Training Data Set*

Characteristic	n
Sex	
Men	126 (51.4%)
Women	119 (48.6%)
Age	
Mean	64.9 ± 12.8 (±sd years)
Median	67
Range	24–98
Delayed-type hypersensitivity skin-test response	
Reactive	155 (63.3%)
Relatively Anergic	46 (18.8%)
Anergic	44 (18.0%)
Presence of Cancer	
Cancer	188 (76.7%)
No Cancer	57 (23.3%)
Surgical Procedures	
Esophagus	10
Stomach	39
Billiary Tree	20
Liver	7
Pancreas	5
Small Bowel	12
Large Bowel	105
Rectum	36
Miscellaneous	11

marized in Table 1. There were 126 men (51.4%) and 119 women (48.6%). One hundred and eighty-eight (76.7%) of the patients were operated on for removal of a cancer of the gastrointestinal tract, and 57 patients had noncancer operations, mostly for diverticular disease of the colon. Major sepsis occurred in 22 patients (9%) with 6 bacteremias, 6 intra-abdominal infections (abscesses or peritonitis), and 10 combinations of intra-abdominal infection with bacteremias. There were 14 deaths for a mortality rate of 5.7%. Twelve of the 14 deaths were clearly due to sepsis or sepsis was contributory, i.e., a major sepsis episode occurred before the patient's subsequent death, but the immediate cause of death was not clearly attributed to sepsis by the attending surgeon or the pathologist. A brief case history of all the patients who died follows.

Case 1

A 72-year-old man presented with dyspepsia. Esophagogastroduodenoscopy demonstrated an adenocarcinoma of the gastroesophagial junction. He was relatively anergic on admission with a DTH score of 9 mm and a serum albumin of 39 g/L. Esophagogastrectomy was done and six days after surgery he showed signs of sepsis and he was immediately explored and found to have a perforated colon with pelvic abscess. He was treated with total colectomy and iliostomy plus drainage of the abscess. Broad spectrum empiric antibiotic therapy in the form of netilmicin, metronidazole and penicillin G was given. He deteriorated with subsequent cardiac arrest three days later. Autopsy revealed a bronchopneumonia, a large subphrenic abscess, residual peritonitis, brain infarction, and pulmonary thromboembolism. The cause of death was stated as heart failure secondary to bronchopulmonary pneumonia and intra-abdominal infection.

Case 2

A 74-year-old adult-onset diabetic man with dysphagia secondary to a carcinoma of the esophagus was admitted with relative anergy, a DTH score of 6 mm, and a serum albumin level of 41 g/L. Esophagogastrectomy (anastomosis in the neck), pyloroplasty, and feeding jejunostomy were carried out. Three days after surgery he developed a temperature of 39 C, and an aspiration pneumonia was diagnosed by chest x-ray and sputum growth of *Enterobacter* species. He was treated with Netilmicin and Clindamycin plus erythromycin (due to an epidemic of Legionella pneumonia in the hospital). Two days later he spontaneously drained a neck abscess with *Enterobacter cloacea* identified on culture. Netilmicin was changed to Tobramycin with pharmacokinetic dosage adjustment and monitoring. He developed hypoxia, obtundation, and a bacteremia with *Seratia marcences* five days later. At this time *Seratia marcences* also grew on sputum culture. Despite aggressive respiratory care and adequate serum levels of tobramycin, he deteriorated and died from gram-negative septic shock, and respiratory and cardiac decompensation.

Case 3

A 76-year-old woman with adenocarcinoma of the sigmoid colon was relatively anergic on admission with a DTH score of 6 mm and a serum albumin of 31 g/L. A Hartman's colonic resection was done as she was thought to be a high-risk patient. She deteriorated over the next five days with bronchopneumonia, and despite broad spectrum empiric antibiotic therapy she developed renal failure followed by hepatic decompensation and death.

Case 4

A 76-year-old man with a solitary metastatasis of colonic cancer to the right lobe of the liver was anergic on admission with a DTH score of 0 mm and a serum albumin level of 13 g/L. He received a right hepatic lobectomy. He was well for 13 days after surgery at which point he became progressively short of breath with reduced urine output. He was treated in the SICU with fluids and Netilmicin plus cefazolin for the suspicion of a hospital-acquired pneumonia. This was confirmed when the sputum grew *Pseudomonas aeruginosa* and *Staphilococcus epidermidis*, and on the following day *Pseudomonas aeruginosa* bacteremia was noted. Vancomycin was added to the antibiotic management. He continued to deteriorate and died. Autopsy disclosed bronchopneumonia and pulmonary hemorrhage at the site of a Swan-Ganz catheter with bacteremia as *Pseudomonas* was grown from the pre- and immediately postmortem blood cultures. Cause of death was respiratory failure.

Case 5

A 63-year-old woman with epigastric pain and a weight loss equivalent to 12% of her body weight had ultrasonographic findings compatible with the diagnosis of pancreatic cancer. She was anergic on admission with a DTH score of 0 mm and a serum albumin level of 31 g/L. The diagnosis of unresectable pancreatic cancer was made on laparotomy and a cholecystectomy, choledochoduodenostomy, and gastrojejunostomy was done. She had extensive intraperitoneal tumor seedings. She deteriorated rapidly after surgery with massive tumor ascites and shortness of breath aggravated by a bronchopneumonia diagnosed by radiologic confirmation of an infiltrate in the lungs and a sputum culture of *Enterobacter species*. She died 10 days after her surgery. Permission for

autopsy was refused by next of kin, and the attending surgeon stated that the cause of death was carcinomatosis with respiratory deterioration.

Case 6

A 62-year-old man with a gastric adenocarcinoma was relatively anergic on admission with a DTH score of 8 mm and a serum albumin level of 27 g/L. He underwent a subtotal gastrectomy and cholecystectomy. On the second postoperative day he developed peritonitis and was taken to surgery. A peripancreatic fluid collection was found that cultured *Staphylococcus epidermidis.* He had three subsequent explorations for intra-abdominal infection, mostly interloop abscesses and peritonitis growing mixed organisms. Despite this and multiple courses of broad-spectrum antibiotic therapy, he deteriorated, developed *Staphylococcus aureus* bacteremia, and died. Autopsy showed generalized peritonitis, pus draining from the ampula of vater, bilateral bronchopneumonia, and pulmonary emboli. Cause of death was stated as intractable septic shock.

Case 7

A 73-year-old man was operated on for resection of an abdominal aortic aneurysm, but at surgery he was found to have a pancreatic cancer. He was closed and discharged with conservative therapy. Four months later he was readmmitted with nausea, vomiting, and a loss of 13% of his usual body weight. He was anergic with DTH score of 0 mm, and a serum albumin level of 30 g/L. A palliative gastrojejunostomy bypass was done. Extensive peritoneal tumor seedings were noted. He developed urinary retention, metabolic acidosis, and a bacteremia with *Staphylococcus aureus* at the same time that the urine grew *Staphylococcus aureus* at 10^8 colony-forming units/ml. He was treated with trimethoprim-sulfamethoxazole intravenously. The patient deteriorated rapidly and died three days later. Permission for autopsy was refused. The death was classified as sepsis related.

Case 8

A 78-year-old man was admitted with pruritus, jaundice, pale stools, and dark urine. He was reactive with a DTH score of 31 mm and a serum albumin level of 32 g/L. At surgery he was found to have a cholangiocarcinoma and a Whipple's resection was done. He was reoperated on within 20 hours because of intra-abdominal bleeding with evacuation of 2 L of clotted blood from the abdominal cavity and control of the bleeding with abdominal packs. These were removed at a third exploration 24 hours later. He developed a wound dehiscence two days after this procedure, and at laparotomy diffuse peritonitis was found. Repeat skin testing at this time showed a DTH score of 0 mm. He was treated with tobramycin and clindamycin and ticarcillin because peritoneal cultures grew *S. aureus, Ps. aeruginosa,* and *Enterococcus.* He subsequently developed an aspiration pneumonia, followed by multiple-organ failure and an *S. aureus* bacteremia. He was operated on for the fourth time and a total dehiscence of the pancreaticojejunostomy was found with retroperitoneal necrosis and abscess formation. He became asystolic during this procedure and he died. No autopsy was requested because it was believed that the laparotomy gave sufficient information. This death was thus classified as sepsis related.

Case 9

A 71-year-old man with an extensive colon cancer was anergic on admission with a DTH score of 0 mm and a serum albumin level of 26 g/L. He was found to have a right colon cancer with large periaortic node involvement and dilated common bile duct. He had a palliative procedure in the form of a choledochoduodenostomy and iliocolostomy. He deteriorated six days after operation with fever, ileus, elevated circulating white cell count, and anemia. He was treated with blood transfusions and antibiotics but he deteriorated rapidly and died. Autopsy showed residual peritonitis with fluid that grew both facultative aerobes as well as anaerobes. Anastomotic sites were intact. Bronchopneumonia was also present. Cause of death was attributed to multiple-organ failure secondary to peritonitis and bronchopneumonia.

Case 10

A 72-year-old man with a gastric cancer was anergic on admission with a DTH score of 0 mm and a serum albumin level of 27 g/L had a subtotal gastrectomy and transverse colectomy due to tumor growth into the transverse mesocolon. He developed peritonitis seven days after surgery with *Klebsiella spp.* He was explored, the abdomen was lavaged, and cefoxitin was given. He continued to deteriorate and developed a right subphrenic and right paracolic abscess diagnosed by CT scan and drained percutaneously. This grew *Ps. aeruginosa* and *Klebsiella pneumonia.* Tobramycin was added to his antibiotic management but he deteriorated and died three days later. Death was attributed to tumor and peritonitis.

Case 11

A 77-year-old woman with pancreatic cancer was relatively anergic on admission with a DTH score of 9 mm and a serum albumin level of 37 g/L. She was treated with a gastrojejunostomy and choledochojejunostomy due to unresectable cancer of the pancreas. She developed an acute hemorrhage from the choledochojejunostomy and was reoperated on for control of this hemorrhage. This was followed by peritonitis and bronchopneumonia, disceminated intravascular coagulation, and multiple-organ failure. She died eight days after operation.

Case 12

An 80-year-old woman was admitted with abdominal pain, nausea, vomiting, and a weight loss 23% of her usual body weight. She was anergic with a DTH score of 0 mm and a serum albumin of 30 g/L. She was diagnosed as having severe diverticular disease with subacute obstruction and had a Hartman's resection. She developed postoperative complications with multiple-organ failure without evidence of major sepsis. She died from congestive heart failure. Autopsy revealed bronchopneumonia with the stated cause of death being respiratory failure secondary to ARDS and bronchopneumonia.

Case 13

A 66-year-old man with a 9% body-weight loss was found to have rectal cancer by rectal exam. He was anergic on admission with a DTH score of 0 mm and a serum albumin level of 26 g/L. He was treated with an abdominoperineal resection. He developed a wound dehiscence eight days after surgery that was repaired. Following this he deteriorated with *Pseudomonas* bacteremia and a wound dehiscence. This was repaired but he deteriorated and died. Autopsy revealed extensive metastatic cancer to the lungs, mediastinum, and the right adrenal gland, with extensive bronchopneumonia. The cause of death was stated as respiratory failure secondary to chronic obstructive lung disease, tumor metastasis, and bronchopneumonia. This was classified as a septic-related death.

Case 14

A 68-year-old man with an adenocarcinoma of the stomach was admitted with skin-test anergy and a DTH score of 3 mm. His serum

TABLE 2. *The Clinical Outcome of the Training Patient Data Set Based on Admission Delayed Type Hypersensitivity Skin-Test Response*

Skin-Test Response	n	Major Sepsis*	Deaths†
Reactive	145	6 (4.1%)	1 (0.7%)
Relative anergy	46	9 (19.5%)	6 (13.4%)
Anergy	44	8 (18.1%)	7 (15.9%)

* $\chi^2 = 16.8$ 2 d.f. $p < 0.0002$.
† $\chi^2 = 20.5$ 2 d.f. $p < 0.0001$.

albumin level was 28 g/L. He had a gastrectomy and resection of a solitary liver metastasis. He developed hemorrhage within 16 hours after surgery and was explored to control the bleeding. He deteriorated three days after this with Gm-negative bacteremia, developed progressive multiple-organ failure, and died 17 days after surgery. Autopsy showed changes in the lung compatible with the adult respiratory distress syndrome and bronchopneumonia, resolving peritonitis, and a urinary tract infection.

Table 2 shows the clinical outcome of the patients according to their skin test response. There were 145 reactive patients (59.1%), 46 relatively anergic patients (18.8%), and 44 anergic patients (17.9%). Both major sepsis ($X^2 = 16.8$, $p < 0.0002$) and deaths ($X^2 = 20.5$ $p < 0.0001$) increased significantly in the nonreactive patients. The data also show the inability of the anergic patient to control major life-threatening sepsis and survive. Most (87.5%) of the anergic patients who developed major sepsis died from it, compared to only 16.7% of the reactive patients ($p < 0.05$). The relatively anergic patients fell in between these two groups at 66%.

Analysis of variance showed that the APS score (without the age points), the age, the polymorphonuclear neutrophil count, serum IgA, and the serum C-reactive protein were significantly increased in the anergic patients, whereas the DTH score, the circulating monocyte count, the serum albumin level, and the hemoglobin concentration were significantly decreased in the anergic patients. The rest of the variables were not significantly different among the three patient groups as shown in Table 3. Table 4 includes the univariate data analysis according to whether the patient survived or died. The DTH score, the APS (minus the age points), patient age, and serum albumin and hemoglobin levels were significantly different among these groups.

TABLE 3. *Analysis of Variance of the Measured Variables According to the Skin-Test Response (mean ± sd)*

Variable	Reactive	Rel Anergy	Anergy
DTH	32.4 ± 14.8	10.1 ± 5.7†	0.7 ± 1.5*
APS1	1.4 ± 1.8	2.0 ± 1.8	2.3 ± 2.3*
Age	61.5 ± 12.5	68.2 ± 12.3†	73.3 ± 9.1*
PMN	4.7 ± 1.7	4.9 ± 2.0	5.6 ± 3.3*
Monocytes	0.51 ± 0.20	0.54 ± 0.23	0.59 ± 0.23*
IgA	2.66 ± 1.15	2.63 ± 1.28	3.50 ± 1.83*
CRP	1.4 ± 2.4	1.0 ± 2.6	3.4 ± 5.7*
Albumin	37.3 ± 5.1	35.7 ± 7.2	33.2 ± 5.8*
Hb	131 ± 20	129 ± 22	115 ± 20*
ADH	65.9 ± 14.6	67.7 ± 14.7	65.1 ± 18.1
CTX	17.9 ± 8.3	16.4 ± 5.8	16.8 ± 8.1
Lymphocytes	1.7 ± 6.3	1.6 ± 6.3	1.6 ± 0.6
IgG	11.9 ± 3.8	12.0 ± 3.6	12.1 ± 3.6
IgM	1.68 ± 2.65	1.38 ± 1.02	1.51 ± 1.54
Total Protein	66.6 ± 7.0	65.9 ± 10.4	64.4 ± 8.7
C3	0.99 ± 0.36	0.89 ± 0.37	0.98 ± 0.35
CH50	35.0 ± 17.9	34.7 ± 16.1	34.6 ± 17.0

* $p < 0.05$ by Bonfferoni corrected t-tests vs reactive patient group.
† $p < 0.001$ by Bonfferoni corrected t-tests vs reactive or anergic group.

TABLE 4. *The Univariate T-Test Analysis of the Variables Between the Patients Who Survived and Those Who Died (mean ± sd)*

Variable	Dead	Alive	p Value
DTH	5.67 ± 8.1	23.6 ± 17.9	0.00001
APS	2.6 ± 1.5	1.7 ± 1.9	0.04
Age	72.0 ± 5.5	64.4 ± 13.0	0.0002
Serum albumin	29.9 ± 6.7	36.7 ± 5.6	0.003
Hemoglobin	112 ± 23	129 ± 21	0.022
Serum CH50	27.1 ± 12.5	35.4 ± 17.6	0.034
Adherence	66.6 ± 22.0	66.0 ± 14.9	NS
Chemotaxis	16.5 ± 7.2	17.3 ± 7.9	NS
PMN	5.8 ± 2.8	4.8 ± 2.1	NS
Lymphocytes	1.5 ± 5.8	1.7 ± 6.3	NS
Monocytes	0.64 ± 0.23	0.52 ± 0.20	NS
IgG	12.0 ± 2.8	11.9 ± 3.8	NS
IgA	2.84 ± 8.9	2.80 ± 1.38	NS
IgM	1.43 ± 1.28	1.60 ± 2.30	NS
C-reactive protein	1.7 ± 3.8	1.6 ± 3.3	NS
Total proteins	60.8 ± 1.26	66.3 ± 0.76	NS
Serum C3	89.3 ± 25.8	97.8 ± 37.1	NS

Logistic regression analysis showed that only two variables significantly and independently contributed to the estimate of the probability of death in the training set. The analysis is shown in Table 5. Previous studies showed an exponential relation between incidence of sepsis and DTH score,[26] thus a logarithmic transformation of the DTH score was done for this analysis, setting DTH scores of 0 to 0.1 mm. The serum albumin level was the "strongest" variable followed by the delayed type hypersensitivity skin test response. The following equation was used to estimate the probability of septic-related death in a patient before operation given the serum albumin level and the delayed type hypersensitivity skin test response.

$$P|\text{death}| = 1/\{1 + e^{(-3.45+1.75*(\text{albumin})+0.3*(\ln[\text{DTH score}]))}\} \quad (2)$$

The shape of the probability of death curve as both variables change from their best to their poorest value is shown in Figure 1. The contribution of each single variable to the probability of death can be determined by holding one variable constant about its mean and changing the other variable from its best to its worst value. This gives a better indication of the significance of abnormalities of these variables compared to the odds ratio because these are continuous and not categorical variables. The contribution of the DTH response is shown in Figure 2, and that of the serum albumin is shown in Figure 3.

This equation was used to calculate the probability of death of a validation data set of patients before operation. Their demographic characteristics and type of surgery is shown in Table 6 Their clinical outcome by delayed type hypersensitivity skin test response is similar to the training data set (Table 7) The overall mortality rate was 5%. Again, reactive

TABLE 5. *The Logistic Regression Analysis of the Training Data Set*

Variable	Coefficient	s.e.	Coefficient/s.e.
Constant	3.45	1.73	1.99
Albumin	−1.75	0.56	−3.16
DTH	−0.30	0.12	−2.55

s.e., standard error.

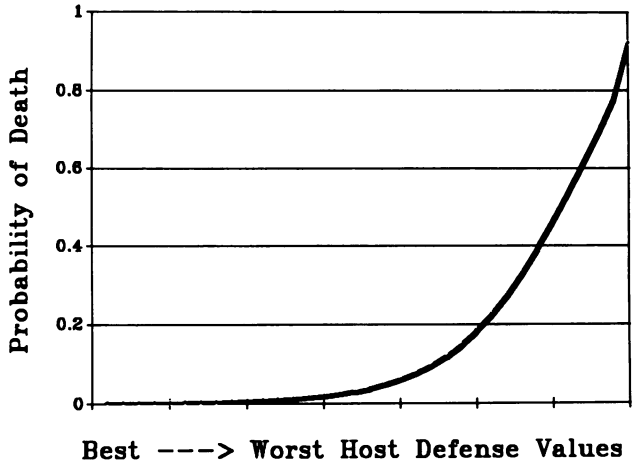

FIG. 1. The theoretical probability of death curve as the independent variables, delayed type hypersensitivity skin test response (99 to 0.1 mm), and serum albumin (55 to 10 g/l) are varied from their best to their worst values.

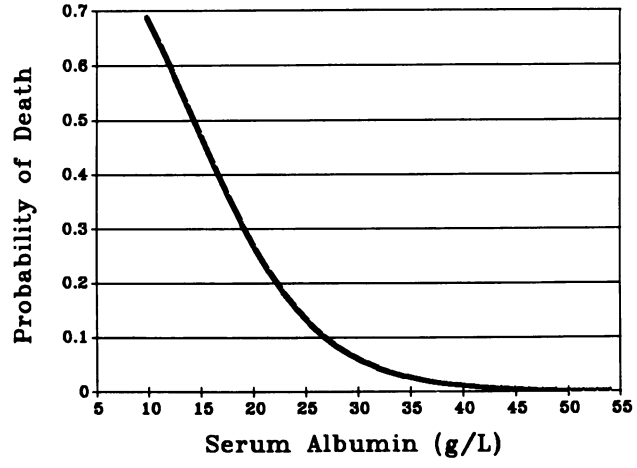

FIG. 3. The contribution of the serum albumin test to the probability of death estimate. Delayed type hypersensitivity skin test response is held constant at 22 mm and serum albumin level varies from 10 to 55 g/L.

patients were able to control major sepsis and survive better then anergic patients (24% *vs.* 75%). Figure 4 shows the expected and observed mortality rates by deciles in this validation set. Overall, 28.3 deaths were expected and 25 were observed. None of the differences are significant (Chi square = 0.139, NS). The Goodman-Kruskell G statistic is 0.66, which indicates a very good correlation between the rank of the patients and the outcome that was actually observed.

The ROC curves and the area under each curve, A_Z, is shown in Figure 5 A_Z for the training set is 0.83 ± 0.06 and for the validation set 0.79 ± 0.05, which indicates a good performance of the probability of death logistic equation.

Discussion

This study confirms the previous observations of many laboratories concerning the ability of the delayed type hypersensitivity skin test response to identify patients at risk for increased septic-related death. In a well-defined preoperative patient population anergy is associated with increased age, increased acute physiology scores, monocyte counts, serum IgA levels, CRP levels, and low serum albumin and circulating hemoglobin levels. When the interactions among these different variables are examined

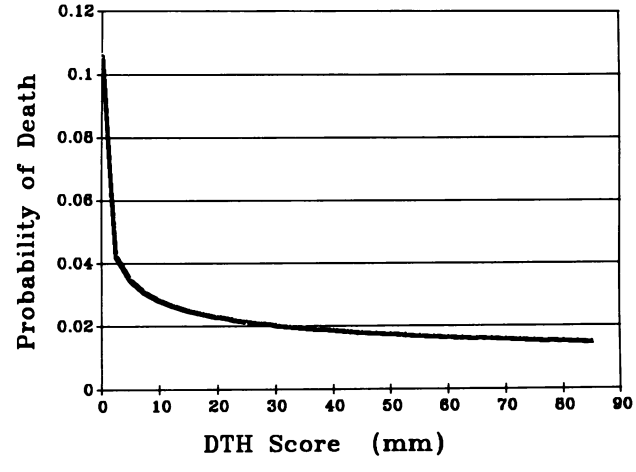

FIG. 2. The contribution of the delayed type hypersensitivity skin test response to the probability of death estimate. The serum albumin level is held constant at 36 g/L and the delayed type hypersensitivity skin test response varies from 0.1 to 90 mm.

TABLE 6. *Demographic Characteristics and Type of Surgery of the Validation Data Set*

Characteristics	Number
Sex	
Men	265 (51%)
Women	255 (49%)
Age	
Mean	64.6 ± 13.0 (±sd years)
Median	66
Range	18–98 years
Delayed-type hypersensitivity skin-test response	
Reactive	355 (68.2%)
Relatively Anergic	77 (14.8%)
Anergic	88 (16.9%)
Presence of cancer	
Cancer	390 (75%)
No Cancer	130 (25%)
Surgical procedures	
Esophagus	22
Stomach	86
Billiary Tree	41
Liver	15
Pancreas	10
Small Bowel	21
Large Bowel	224
Rectum	75
Miscellaneous	26

sd, standard deviation.

TABLE 7. *The Clinical Outcome of the Validation Patient-Data Set Based on Admission Delayed-Type Hypersensitivity Skin-Test Response*

Skin-Test Response	n	Major Sepsis	Deaths
Reactive	355	33 (9.3%)	10 (2.8%)
Relative anergy	77	6 (7.8%)	6 (7.8%)
Anergy	44	14 (15.9%)	10 (11.3%)

by logistic regression analysis, only delayed type hypersensitivity skin test response and serum albumin levels significantly and independently affect the outcome variable of interest, i.e., septic-related death.

This observation is not explained on the basis of abnormalities of nonspecific immunity as reflected by numbers and *in vitro* function of polymorphonuclear neutrophils. These cells are the first line of defense against bacterial invasion. It remains to be shown whether an adequate number of such cells is delivered to areas of bacterial invasion in anergic patients. If not, such reduced polymorphonuclear neutrophil delivery may allow for bacteria to establish a "beach head" from whence they may propagate and form a local abscess or, along with abnormalities in specific immunity due to the "anergic environment" of the host, may progress to systemic invasive sepsis. We have shown previously that lymphocytes do not work properly *in vivo* in an anergic host, although they have the capability for normal responses when stimulated *in vitro*. We have also shown that anergic surgical patients have a reduced ability to generate an antibody response to protein antigens, although their antibody response to polysacharide antigens is normal. Studies of polymorphonuclear neutrophil delivery and function to inflammatory sites by use of blister skin windows are ongoing.

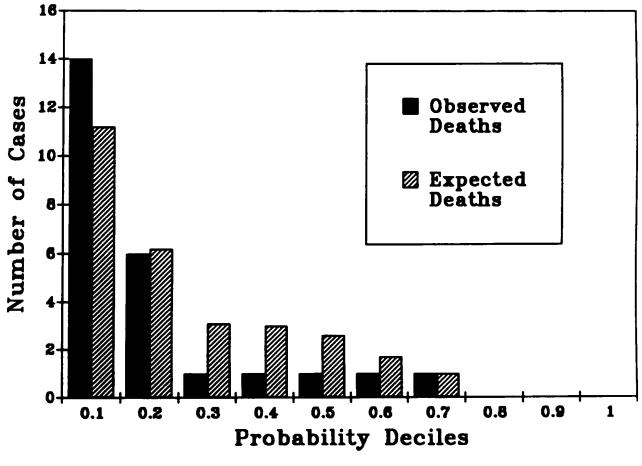

FIG. 4. The expected and observed deaths within each probability decile.

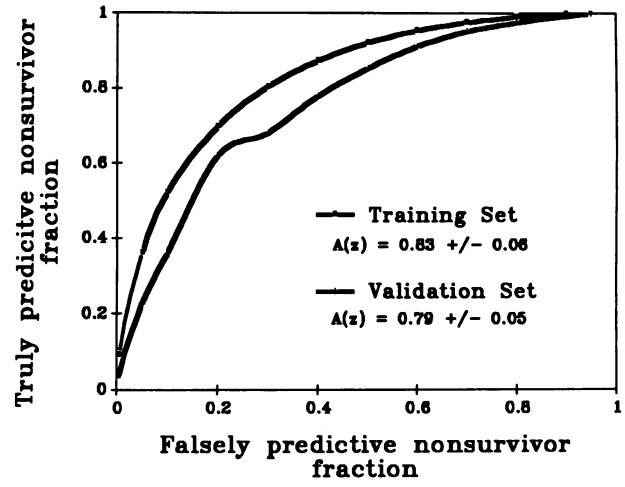

FIG. 5. Receiver-Operating-Characteristic curves for the training and validation data sets. A_z indicates area index.

When factors of age, type of surgery, serum albumin, hemoglobin levels, circulating cells, and acute-phase protein responses are controlled for, the delayed type hypersensitivity skin test response independently and significantly contributes to the risk assessment of the patient before operation. This is evidence against the hypothesis that anergy reflects the nutritional status of a patient. There was a highly significant direct correlation between the delayed type hypersensitivity skin test response and serum albumin ($p<0.001$) levels. When this was controlled for in the logistic regression analysis, both these variables independently contributed to an adverse outcome probability calculation. If the delayed type hypersensitivity skin test response measured malnutrition alone, as some authors have suggested,[27] then it should not have independently contributed to the probability of mortality calculation.

The serum albumin is the other variable of those measured that also contributes to this score. What the serum albumin test measures is open to speculation. It would be simple to attribute the low serum albumin level to malnutrition, notably in view of the weight loss of most of the anergic subjects who died. However there were a proportionately equal number of reactive patients with low serum albumin levels and weight loss who were operated on and tolerated their operation extremely well. The mean serum albumin level was lower in the anergic patients compared to reactive patients but this value was still within the 95% confidence limits of the measurement and greater than the usually accepted level for malnutrition, i.e., 32 g/L. The serum albumin level and C-reactive protein were inversely correlated. Another plausible explanation of these findings is that the reduced serum albumin level indicates a "down regulation" of albumin

synthesis and an "up regulation" of acute-phase response proteins such as CRP. This "condition" exists in some patients due to a systemic event, e.g., response to their surgical disease, which may or may not be beneficial to survival.

Another interesting observation is the lack of contribution of the patients' age to the mortality-risk equation. It would appear that the delayed type hypersensitivity skin test response or the serum albumin test, or both, measure the mortality risk indicated by chronologic age. As long as one is reactive with a normal serum albumin level, then the chronologic age is irrelevant to this risk analysis.

This approach can identify patients at increased risk for septic-related death. Its clinical application is limited by the precision of the measurement. The G statistic is 0.67, and ROC analysis gives an A_z of 0.79. The ideal risk index would have both these measurements at 1. A lack of corrective measures to the defects adds to this limitation. An important question raised by the data is the role of preoperative nutritional support in patient outcome. It is now possible to give enteral or parenteral nutritional support that will correct the low serum albumin level of these patients. If the increased risk derives from malnutrition alone, then one should see a marked improvement in high-risk patients. This approach allows the identification of such a high-risk patient group that can be used in modulation studies, such as preoperative parenteral or oral supplementation that influences both immune and nonimmune antibacterial host defenses, immunomodulating drug regimens, and so on. When data from studies such as the VA cooperative trial of perioperative total parenteral nutrition[28] and others similar studies become available, they may provide means for physicians to adequately prepare their high-risk patients for needed surgical intervention. Until such studies are done, the risk analysis reported here is useful mainly in the research laboratory to identify the appropriate patients to be studied in order to elucidate the reasons for the increased mortality risk.

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