a new source of agents for passive immunization, monoclonal antibodies. Using murine monoclonal antibacterial antibodies, promise in prophylaxis against sepsis has been shown in animal models.[28] The blood lymphocytes spontaneously secreting antibody may be a useful source of cells for the production of human monoclonal antibodies, provided the kinetics of *in vitro* antibody production are understood.

In conclusion, we have demonstrated decreased *in vivo* and *in vitro* humoral immunity in a heterogenous group of surgical patients. There is a kinetic and quantitative correlation between *in vivo* and *in vitro* responses, the latter being a biologic reflection of the magnitude and integrity of the *in vivo* process. Failure to produce specific antibody is not due to failure of total IgG synthesis. Although there will be important new applications for active immunization against bacteria, further knowledge of humoral immune processes in man is required to facilitate exploitation and modulation of *in vivo* responses. The development of human monoclonal antibacterial antibodies for passive immunization will be facilitated by increased understanding of *in vitro* humoral immune responses.

## References


1. Young LS, Stevens P, Kaijser B. Gram-negative pathogens in septicaemic infections. Scand J Inf Dis 1980; 31(S):78–95.
2. Stamm WE, Martin SM, Bennett JV. Epidemiology of nosocomial infections due to gram-negative bacilli: aspects relevant to development and use of vaccines. J Infect Dis 1977; 136(S):151–160.
3. Kreger BE, Craven DE, Carling PC, McCabe WR. Gram-negative bacteremia. III. Reassessment of etiology, epidemiology, and ecology in 612 patients. Am J Med 1980; 68:332–343.
4. Zinner SH, McCabe WR. Effects of IgM and IgG antibody in patients with bacteremia due to gram-negative bacilli. J Infect Dis 1976; 133:37–45.
5. MacLean LD, Meakins JL, Taguchi K, et al. Host resistance in sepsis and trauma. Ann Surg 1975; 182:207–217.
6. Meakins JL. Clinical importance of host resistance to infection in surgical patients. Advances in Surgery 1981; 15:225–255.
7. Christou NV. Host defense mechanisms in surgical patients: a correlative study of the DTH skin test response, granulocyte function, and sepsis in 2202 patients. Submitted to Ann Roy Coll Surg, Canada, 1984.
8. Mazaheri R, Rode HN, Abikar K, et al. Dysfunction of humoral immunity in anergic surgical patients: absence of anti-tetanus IgG antibody production. J Clin Immunol 1984; 4:65–70.
9. Nohr C, Christou NV, Broadhead M, Meakins JL. Failure of humoral immunity in surgical patients. Surg Forum 1983; 34:127–129.
10. Stevens RH, Saxon A. Immunoregulation in humans. Control of antitetanus toxoid antibody production after booster immunization. J Clin Invest 1978; 62:1154–1160.
11. Markwell MAK, Fox CF. Surface specific iodination of membrane proteins of viruses and eucaryotic cells using 1,3,4,6-tetrachloro-3a,6a-diphenylglycoluril. Biochemistry 1978; 17:4807–4817.
12. Christou NV, Meakins JL. Phagocytic and bactericidal function of polymorphonuclear neutrophils from anergic surgical patients. Can J Surg 1982; 25:444–448.
13. Christou NV, Meakins JL. Neutrophil function in surgical patients: two inhibitors of granulocyte chemotaxis associated with sepsis. J Surg Res 1979; 26:355–364.
14. Superina RA, Christou NV, Meakins JL. Failure of neutrophil delivery into skin windows in anergic surgical patients. Surg Forum 1980; 31:68–70.
15. Meakins JL, Pietsch JB, Bubenick O, et al. Delayed hypersensitivity: indicator of acquired failure of host defenses in sepsis and trauma. Ann Surg 1977; 186:241–250.
16. Rode, H, Christou NV, Bubenik O, et al. Lymphocyte function in surgical patients. Clin Exp Immunol 1982; 47:155–161.
17. Stevens RH, Macy E, Morrow C, Saxon A. Characterization of a circulating subpopulation of spontaneous antitetanus toxoid antibody producing B cells following *in vivo* booster immunization. J Immunol 1979; 122:2498–2504.
18. Falkoff RJ, Zhu L, Fauci AS. Kinetics of the *in vivo* activation of antigen-specific B lymphocytes following booster immunization with tetanus toxoid. Clinical Research 1982; 30:347A.
19. Nohr CW, Christou NV, Rode H, et al. *In Vivo* and *in vitro* antibody responses to tetanus toxoid in man. Fed Proc 1984, in press.
20. Benner R, Hijmans W, Haaijman JJ. The bone marrow: the major source of serum immunoglobulins, but still a neglected site of antibody formation. Clin Exp Immunol 1981; 46:1–8.
21. Meakins JL, Nohr CW. Assessment of immunologic responsiveness. *In* Kirkpatrick JR, ed. Nutrition and Metabolism in the Surgical Patient. New York: Futura Publishing Company, 1983:107–137.
22. Bistrian BR, Blackburn GL, Hallowell E, Heddle R. Protein status of general surgical patients. JAMA 1974; 23:858–860.
23. Twomey P, Ziegler D, Rombeau J. Utility of skin testing in nutritional assessment: a critical review. JPEN 1982; 6:50–58.
24. Nohr CW, Meakins JL. Biological response modifiers. In: Meakins JL, ed. Surgical Infection in Critical Care Medicine. London: Churchill Livingstone, 1984, in press.
25. Jones RL, Roe EA, Gupta JL. Controlled trial of *Pseudomonas* immunoglobulin and vaccine in burn patients. Lancet 1980; ii:1263–1265.
26. Zeigler EJ, McCutchan A, Frier J, et al. Treatment of gram-negative bacteremia and shock with human antiserum to a mutant *Escherichia coli*. New Engl J Med 1982; 307:1225–1230.
27. Kohler G, Milstein C. Continuous cultures of fused cells secreting antibody of predefined specificity. Nature 1975; 262:495–497.
28. Dunn DL, Mach PA, Cerra FB. Monoclonal antibodies protect against lethal effects of gram-negative bacterial sepsis. Surg Forum 1983; 34:142–143.


---

### Discussion

DR. LOREN J. HUMPHREY (Shawnee Mission, Kansas): Dr. Nohr and colleagues have stratified surgical patients on the basis of response to skin test antigens and found that specific production of antitetanus toxoid antibody *in vitro* and *in vivo* correlated with the skin testing. Thus, humoral immunity to a specific antigen was depressed at a time when the general immunoglobulin level was normal.

In 1975, Slade, Simmons, and co-workers measured 12 parameters of immune function and found that all *in vitro* functions studied, such as total peripheral blood lymphocyte count, B-cell count, T-cell count, and lymphocyte blastogenesis to mitogens, fell with induction of anesthesia and continued to fall during and after surgery. Thus, cell immunity is depressed as well.

In 1970, Dr. Frederickson and I showed that anesthesia and high levels of oxygen *in vitro* caused a decrease in production of antitetanus toxoid antibody. Interestingly, these could be rejuvenated appropriately *in vitro* to produce antitetanus toxoid antibodies again.

Of further interest to surgeons is the fact that it has been shown that tumor burden as well decreases immune responsiveness.

Why do these various agents cause immunosuppression, and what can we do to prevent this? Certainly, as shown by Dr. Nohr and his

colleagues, this is a double jeopardy for the patient. The patient who begins in a hyporesponsive state then has added on top of this the immunosuppression of anesthesia and surgery.

Two years ago, we reported to the Southern Surgical Association that immunization with an allogeneic tumor protein in cancer patients can stimulate the immune responsiveness, so that it does overcome this immunosuppression. Subsequently, by studying subsets of lymphocytes, we have shown that this immune stimulation is associated with a decrease in T suppressor cells, but, surprisingly, at the same time, a decrease in NK or natural killer cells.

This morning's paper brings to us one of the vital issues of surgeons: How can we overcome immunosuppression? I believe that specific immunologic stimulation of the surgical patient will be the watchword of this decade. To this point, I would ask the authors: What are your plans to determine the mechanism of this specific immunologic suppression?

I think this is a magnificent paper.

PRESIDENT RAVITCH: Thank you, Dr. Humphrey. The discussion will be continued by Dr. John Mannick, of Boston, who walks very well after having his spine replaced by steel.

DR. JOHN A. MANNICK (Boston, Massachusetts): The paper we have just heard is one that I think is important. I think it is particularly important because there is a general belief that, while cellular immunity is depressed in surgical patients, traumatized patients, burn patients, humoral immunity is left intact.

Now, the evidence upon which this assumption is based is probably underwhelming when it is examined closely, and I think that the group at McGill were very correct to test this hypothesis again in their group of anergic patients.

We have not studied a similar group of patients, but have looked at the same phenomenon—that is, the ability to respond with an anamnestic response to tetanus toxoid—in a group of severely burned patients and we have somewhat similar findings. We found, for example, that the mean peak antibody titers were reduced in the burn patients after standard tetanus booster immunization, just as the authors have.

We also found a phenomenon that I am not sure they have encountered, and I would like to ask them about it. We discovered that even in those patients who made a reasonably good response to the antigen, in terms of antitetanus antibody–that the antibody titers peaked out quickly, and then faded fast. In other words, there was no prolonged response, as you have seen in their slides of the normal antitetanus response, but a quick falloff that we find totally inexplicable.

I wonder if they have encountered this phenomenon; and if so, if they have an explanation to offer for it. I have enjoyed the paper very much.

DR. JOSEF E. FISCHER (Cincinnati, Ohio): The excellent presentation by Dr. Nohr represents another of their contributions to the concept of trying to define the population at risk, and specifically as it applies to surgical patients. I think it is fair to say that, although there has been a lot of work in this area, and it has been demonstrated statistically that anergy, or lack of response, perhaps especially in the surgical patient group, correlates statistically with a poorer outcome, it has not been successfully demonstrated, to my knowledge, that in an individual patient anergy statistically relates to changes in outcome.

What we have heard today is, I believe, a major change in direction from what previously has been supposed as being a total failure of responsiveness to something that might better be called dysregulation; in other words, that it is not a total failure of the system, but a failure of the directive system, and that, far from being completely wiped out, if you will, the system works, but does not work very well, and the switch is gone.

Obviously, this is a major contribution, and I would like to ask the authors (1) if they have any concept of where the failure in regulation is; (2) since their approach and our approach over the past decade or so has been, perhaps, belief that nutritional manipulation might help this particular problem, whether or not they have any clues in their very extensive work as to where intervention in a specific way might alter the outcome.

DR. CHARLES E. LUCAS (Detroit, Michigan): Our own data indicate that the decrease in the response to tetanus toxoid after trauma is further increased in patients who have received albumin. Can you relate your experiences about the response of tetanus toxoid to patients who have received albumin *vs.* no albumin?

DR. C. W. NOHR (Closing discussion): Dr. Humphrey, clearly the mechanisms involved between the injection of an antigen or the encounter of a host with an antigen and the eventual production of antibody are complex and poorly understood. For *in vitro* production of specific antibody by unstimulated cultures, recent antigenic priming *in vivo* is required. Although we have no experiments as yet to delineate the exact mechanism of the failure of patients to produce specific antibody, it may occur in the process of antigen recognition, and transfer of this specific information from cell to cell, involving clonal proliferation of specifically sensitized B-cells and the production of intercellular mediators—for instance, interleukin-1 and -2.

Regarding the antibody responses of burn patients, we agree that burn injury probably represents a unique kind of insult to the host, and that unique immunological alterations may be consequent thereto. We do not have human burn data, but our experiments in animal models show that humoral immune responses are blunted.

Dr. Fischer, we agree with the use of the term "dysregulation." The use of the term "total failure" probably reflects our ignorance about some of the aspects of the immune processes involved. Again, as to where the exact location of failure will occur in this dysregulated process, we have not described.

The influence of nutrition is of tremendous interest because it is one of the causes of immune deficiency that we can address therapeutically. It is interesting to us that two of the anergic patients in our study that produced substantial responses to tetanus toxoid had been started on enteral nutrition just days before that. However, there was another patient similarly started on enteral nutrition who failed to respond; so the numbers are obviously too small to reach any conclusions. The hint suggested at by this human data is supported by animal experiments done by other individuals, which suggest a state of immune hyperresponsiveness, soon after the initiation of refeeding in malnourished animals.

We do not have any information on the influence or lack of influence of albumin transfusion on humoral immunity.