IMMUNODEFICIENCY IN MAN AND ANIMALS

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IMMUNODEFICIENCY

IN MAN AND ANIMALS

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IMMUNODEFICIENCY IN MAN AND ANIMALS

INTRODUCTION

The Second International Workshop on the Primary Immunodeficiency Diseases came as an outgrowth of an informal meeting among members of the World Health Organization Expert Committee on Immunodeficiency. Surely the time was at hand for another Workshop and late in the summer of 1972, while attending a Workshop of another subject, Doctors Fred Rosen, Hugh Fudenberg, Robert Good and myself spent an evening together formulating a tentative program of topics to be discussed and participants to be invited. Later Dr. Henry Kunkel was consulted as were Dr. Walter Hitzig, Dr. Maxime Seligmann and Dr. John Soothill, all members of the WHO Expert Committee on Immunodeficiency.

The program began where the first Workshop ended. The results of the first Workshop were published in a book entitled *Immunologic Deficiency Diseases in Man.* The sophisticated delineation of the two-component concept of immunity, and further characterization of the T and B cells and their respective functions and perturbations in disease opened the Second International Workshop on the Primary Immunodeficiency Diseases in St. Petersburg Beach, Florida held February 4-8, 1973. Since 1967, when the first Workshop was held, progress made in numerous laboratories and medical centers throughout the world in diagnosis, analysis and treatment of these fascinating diseases has been great indeed.

By the end of the first Workshop the moment had come when our knowledge and technology made it possible to theoretically formulate the basis for correction of some of these immunodeficiency diseases by cellular engineering. Indeed, the feat was accomplished in 1967 a few short months following the Workshop. Since then improved methodologies for histocompatibility testing and matching have made possible the correction of many cases of inborn immunodeficiencies and it is now theoretically possible to extend this achievement in the years to come. Complete correction of all these deficiencies is a goal to be sought and we have within our command an expanding technology and biologic understanding to make this come true.

Papers and discussions presented at this Workshop showed us that we have entered an era in which biochemical analysis of immunologic development will be possible. This means that ultimately our definitions and analyses of the fundamental biochemical genetics of the abnormalities underlying the immunodeficiencies will permit macromolecular engineering to supplement or even replace the cellular engineering so much on our minds during this Workshop.

Over and over again this Workshop, like the first, revealed how much our knowledge of the normal immunologic functions depends upon the studies of the natural experiments represented by the rare instances of inborn errors in man and animals. The inborn deficiencies of the complement components, for example, more than any in vitro analysis show us the vital importance of the individual complement components and the several complement pathways in survival.

The frequent depressions of immunologic function that occur as a consequence of infection, nutritional deprivation, aging and cancer are being analyzed and understood in the focus made possible by study of the primary immunodeficiencies.

How encouraging is the fact that new names and new personalities played major roles in the second Workshop. The steadying old hands and critical minds were there, but the emphasis was clearly on youth. As is appropriate to such a Workshop, the vigorous discussions left many unanswered questions thereby assuring the need for additional meetings in future years. Surely transfer factor will soon be defined, the biochemical analysis of thymic influence has begun, histocompatibility matching in the general population promises finally to become a reality. New diseases have been defined and the old ones realigned and reassessed and diagnostic screening is now within the reach of clinics throughout the world. Ataxia-telangiectasia and Wiskott-Aldrich syndrome are still puzzles too difficult to handle. But even here new information and new methodologies promise new understanding and ultimately definitive analysis. And finally a series of useful animal models mimicking certain immunodeficiencies will permit more rapid analysis of these diseases in man.

It is possible to look forward to a third Workshop confident that in several more years many of the questions will be answered and new and improved means for treating patients with these challenging diseases will be available. Hopefully, as a spin off from these efforts, new approaches to the management of autoimmune diseases, cancer, infectious diseases, allergy and the aging of the immunity systems can be realized.

Joanne Finstad

WELCOME

It is my pleasure to represent The National Foundation-March of Dimes and to welcome you to this second workshop on the Primary Immunodeficiency Diseases in Man. We were happy to co-sponsor the first workshop in Sanibel Island six years ago. Happily, some of you were there.

The proceedings of that workshop were published and fortunately, or unfortunately as the case may be, every last copy of those proceedings has been sold. We are looking forward to publishing the proceedings of this workshop also.

We welcome you here very sincerely, and it is my personal pleasure to be here, to visit with you again, and to listen to the exciting discussions and fascinating papers that will be presented.

Daniel Bergsma, M.D., Ed.

PREFACE

Although there is nothing on the program about this, I have been given the pleasant task of making a few general comments before the meeting begins. Let me hasten to add, specifically for your benefit Dr. Good since you know nothing about this, that I am not usurping your prerogative. We all know and appreciate what you have done to make this meeting possible. However, much as you might wish to do so, the presentation of these introductory remarks must fall to someone else; the purpose is to dedicate the published volume to you. We realize, of course, that it is a bit unusual to do this for someone we presume will be an active participant. However, the overwhelming opinions prevailed and no one thought that in your case, such an act would in any way suppress such participation. Nor does it absolve you from your duties as an editor of the volume. In fact, in return, we expect a little extra out of you.

It would be superfluous for me to recount Bob Good's accomplishments in the immunodeficiency area, particularly to a group like this, but suffice to say that he is truly the father of immunodeficiency. Perhaps father is the wrong term because that leaves out the Brutons. Perhaps mother would be better; in fact, mother is better for many other reasons! It must be almost 25 years ago that we worked together in New York for a while, and I was caught up in his contagious enthusiasm that I know you all know so well. Even at that early date, his primary interest was in cells, lymphocytes and plasma cells and he almost converted me but I could only see immunoglobulins. Now these cells represent the dominant theme of immunology and I have to recall with admiration such unique foresight. The subject of the lymphocyte will also surely dominate our meetings here.

In addition to Dr. Good's monumental qualifications, there is another reason for this presentation. Now that he is beginning his duties as Director of the Sloan-Kettering Cancer Institute, this meeting probably represents something of a "Swan Song" to his work in immune deficiencies. How many in this audience are naive enough to believe that statement?

Some years ago, at a symposium in Europe, we had some fun at Dr. Good's expense and I served a similar function and gave a serious presentation to him for a "Special Immunopathology Award." It was a defeathered bursectomized rubber chicken. This time, however, it is no joke and I speak very sincerely, on behalf of all of us here, in dedicating this volume to you, Bob.

Henry G. Kunkel, M.D.
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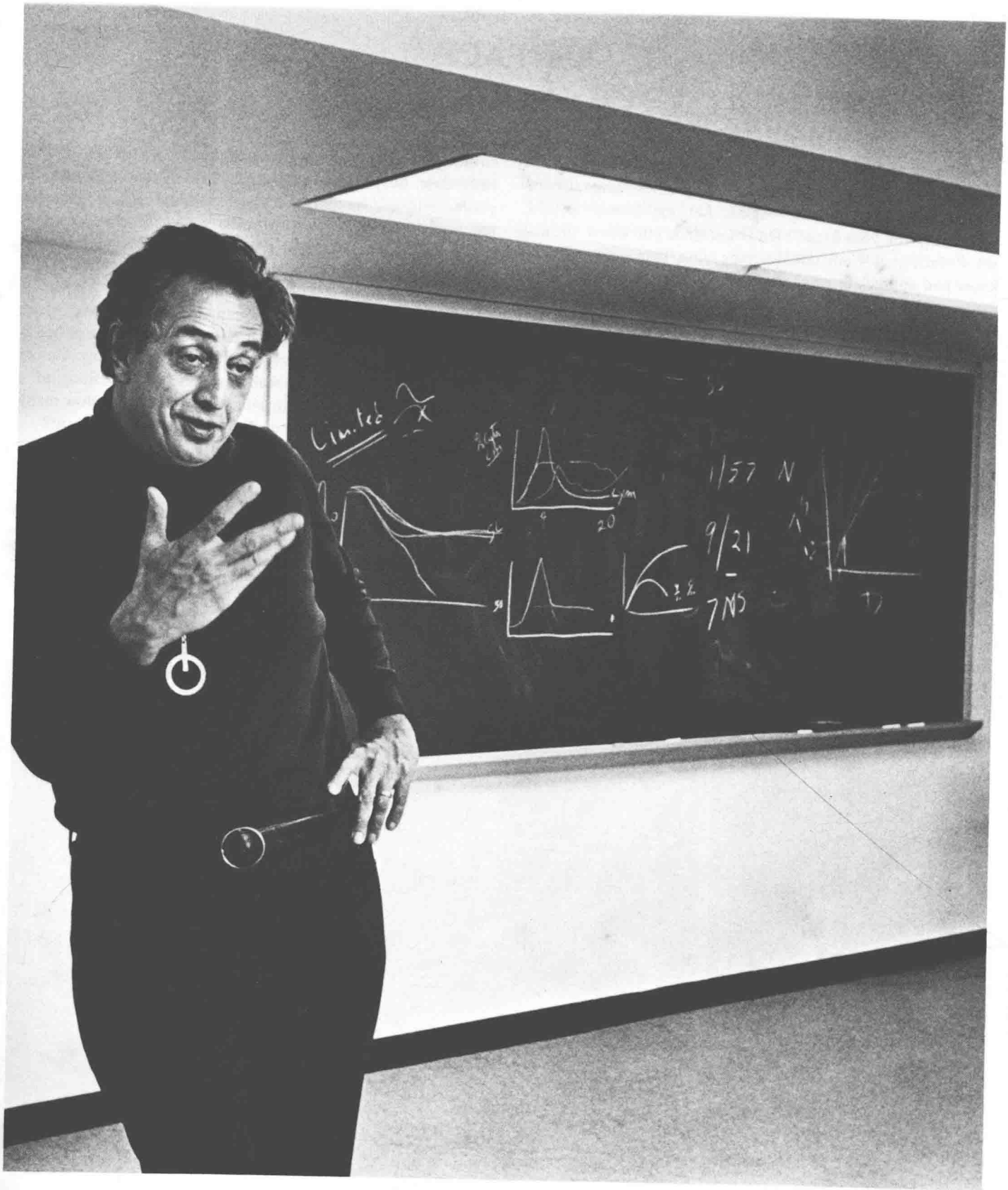
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IMMUNODEFICIENCIES OF MAN AND THE NEW IMMUNOBIOLOGY

ROBERT A. GOOD, M.D., Ph.D.

We had come a long way by the time of the 1967 Sanibel Workshop on Primary Immunodeficiency Disease, but at the conference, as is so often the case, we also found that we had come farther than we had realized. We found that vectors could form from the interaction of the scientists in the field and at the bench and that these vectors could provide direction for developments of the future. In the 6 years since 1967 we have even come further. For some of us, the way has seemed painfully slow, but we can anticipate in planning a third workshop that a logarithmic progression of knowledge and achievement will be apparent. Thus, perhaps we will be ready in 3 years for the next exchange. We come here now 1) to establish where we are at this moment in history, 2) to review what we have been doing, 3) to look into the future and 4) to design the new experiments. We are deeply indebted to The National Foundation for support in this workshop and to Dr. Daniel Bergsma, whose deep concern in the area of birth defects has made this workshop possible.

Man lives in a sea of microorganisms, and he maintains his integrity by being able to defend himself against both external and internal invaders. Man does this by considering these microorganisms to be foreigners, by being able to identify them and eliminate them from his body or by sharply controlling their presence in his body. This is a remarkable achievement, which we know involves the operation of 2 separate specific immunity systems. The latter are comprised of complex interacting networks of cells distributed in blood, lymph, lymph nodes, spleen, marrow and connective tissues. These cells have the job of recognizing what is foreign and by the products of their cells engaging the biologic amplification systems which in turn activate effector mechanisms. We now know that the biologic amplification systems can expand, up to several thousand-fold on a molecule-for-molecule and cell-for-cell basis, the influences of the specific immunity systems. Great progress has been made over the past 2 years in understanding precisely how the specific immunity systems, biologic amplification systems, and effector systems work. Indeed, how they work together to achieve the extraordinary defense of the body. Generation of the new information is often derived from the study of "experiments of nature", in which one or another of the essential components of the bodily defense is missing or deranged.

The "experiments of nature" which I personally consider to have been milestones in the development of present understanding of the immunity systems were 1) the study in 1937 of agranulocytosis by Bing and Plum.[1] These investigators studied 13 cases of agranulocytosis; 3 had both extraordinary hyperglobulinemia and plasmacytosis. They suggested that the plasma cells may be producing the globulin that was accumulating in the blood. 2) The contributions of Fred Kolouch, at the University of Minnesota, 1937 to 45, derived from a study of a patient who died of subacute bacterial endocarditis. Kolouch had noted that plasma cells were abundant in the bone marrow studied at autopsy. From this observation he associated antigenic stimulation and the accumulation of plasma cells. He took the question posed by this experiment of nature to the laboratory and carried out the first experiments in the modern era linking antibody production to antigenic stimulation. I helped Kolouch do certain of his controls and as a result became interested in plasma cells.[2,3] My own role in the Kolouch work was to do 2 things: A) to show that anaphylactic shock, when made possible by passive immunization, did not induce extensive plasma cell development. This finding ruled out the anaphylactic shock reaction per se as the stimulus to the development of these beauiful cells.[4] B) I further showed that secondary stimulation with simple protein antigens, like ovalbumin and bovine serum albumin, could induce plasma cell production just as had the complex bacterial vaccines Kolouch used.[5]

In addition, I carried out rather extensive studies using a variety of tissues including the brain to show that injections of antigens into tissues in previously immunized rabbits regularly led to the prompt differentiation of plasma cells at the local site apparently from antigenic stimulation of the lymphocytes participating in the acute inflammatory reaction induced.[6] As a consequence of these exciting studies, by the time I got to The Rockefeller University, I was already considered to be a plasma-cell hunter, one of that early lineage who considered plasma cells to be antibody-producing cells. I was pleased to find that a number of scientists in New York were also interested in plasma cells. Among them was Henry Kunkel, who wanted to study this issue from the perspective of immunochemistry. He wanted to compare myeloma proteins, one with another and with normal gamma globulins, using immunochemical technics. Perhaps because I was a plasma-cell hunter, it was possible for me to obtain a number of myeloma sera and thus help Henry Kunkel launch what turned out to be the first experiments that led to the unraveling of the chemistry of immunoglobulins, indeed to the complete analysis of antibody structure. Now let me admit here and now, I was not much interested in Kunkel's approach, that history now

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shows turned out so well, but I was more interested in my own musings from clinical encounters with the myeloma patients. You see, I drew blood from some of these myeloma patients and thus had a chance to talk with them and to hear that they often had terrible problems with infections. From their doctors, and from the stories told by the patients, it was clear that their infections were not infections caused by *all* organisms, but were infections with only *certain* organisms. These patients developed pneumonia, and septicemia particularly with pneumococcus, Hemophilus, and Pseudomonas organisms.

At the same time, I was in Maclyn McCarty's department, and wanting to please my mentor, I was determined to crystallize C-reactive protein (CRP). To do this, I tried to find patients with streptococcal empyema that would produce the kind of effusions from which McCarty originally crystallized CRP. By 1949 no such patients were available. Antibiotic treatment of streptococcal infections had dried up that source of pleural fluid. I found, however, that effusions from patients with Hodgkin disease, who often had huge effusions, contained much CRP. Having to obtain pleural effusion fluid from patients with Hodgkin disease, I soon learned that they, too, were abnormally susceptible to infection; but their infections were completely different from those of the myeloma patient. These patients were troubled by recurrent infections with tuberculosis, fungi and viruses. Particularly, they were troubled with herpes and the pox viruses. In addition, other pathogens that we considered to be lower grade bacterial pathogens were causing much trouble. It was in this counterpoint relationship between the organisms plaguing patients with myeloma and Hodgkin disease that concerned me. It was from these early musings that I began to wonder whether there might just be not one single immunity system, but maybe at least separate immunity systems that would be separately perturbed in disease.

Already by 1955, I began to write about this possibility. There is no question but that we received a tremendous assist in this analysis from studying patients with X-linked infantile agammaglobulinemia. These patients, just like the myeloma patients, were troubled by infections almost exclusively attributable to the high-grade encapsulated extracellular pyogenic bacterial pathogens. They could develop delayed allergies but could not produce circulating antibodies.[7-9] It was then that Schier[10] reported that patients with Hodgkin disease lacked delayed allergy. We followed his lead and quickly demonstrated that patients with Hodgkin disease have a very different immunodeficiency from the agammaglobulinemic children or the patients with myeloma.[11-17] They did not develop or express delayed allergic reactions normally, but made antibodies well. They did not reject allografts of skin with vigor, but had normal levels of immunoglobulins. The myeloma patients and agammaglobulinemic patients, by contrast, made antibodies poorly but developed delayed allergy well. They also rejected skin allografts quite well in most instances. Thus, at its very inception the very germs of the two-component concept of the lymphoid system was the function of interpretation of several experiments of nature. We had proposed clearly the concept that 2 separate immunities existed from the implications of these crucial natural experiments long before the concept could formally be stated in terms of different influences of thymus and bursa. Nonetheless the concept was stated again by the experiments of Warner and Szenberg[18] and the clarifying analysis of Cooper and Peterson in my laboratory.[19]

Now we know also, from extensive studies with experimental animals as well as with man, that the T-cell population, via its own products which act as biologic amplification and effector mechanisms, represents in some way a major defense against certain viruses, fungi, or facultative intracellular bacterial pathogens.[20, 21] We even have begun to learn something about the means by which this defense is accomplished; however, we have been very slow to define the molecular basis of T-cell action. We still look on the B cells in a major way as a system for dealing with high-grade encapsulated pyogenic pathogens. It is exciting to reflect more on what we are seeing here. Indeed, for the first time it makes sense to classify together streptococci, Hemophilus influenza bacilli, pneumococci, Pseudomonas aeruginosa and meningococci, on one hand; and certain viruses, many fungi, and tubercle bacilli, Salmonella and Histoplasma for example on the other hand, as representing different microbial universes. In a real way, the immunodeficiencies are helping us to classify the microbial world according to means by which we, as a component of an evolutionary scheme, have learned through the eons of evolutionary history to defend ourselves against the different organisms in the microbial universe.

We now also began to think, very early, in terms of the fundamental effector mechanisms. What are the ways by which these 2 separate immunity systems get their work done? There was of course the inflammatory reaction, but each immunity system seems to use inflammation in a different way. In this context we considered the teachings of Lewis Thomas who always insisted that it is wise to keep each component of inflammation separate and not to lump the many events occurring in inflammation into a single category. The immunity systems, it was apparent, use phagocytosis, vascular reactivity based on smooth muscle contractions, and inflammation, in a variety of ways and even blood coagulation to get their job done.

The 2 immunity systems engage the basic effector mechanisms by means of the biologic amplification systems listed in Table I.

TABLE I

1. Classic complement pathway
2. Alternate or shunt complement pathway
3. Kallekrein – kinin system
4. Release of vasoactive amines by cellular secretion
5. Production by lymphocytes of lymphokines including MIF, MAF, cytotoxins, blastogenic factors, vascular permeability factor, transfer factor and interferon, LIF.

The development of our knowledge concerning the really important aspects of how these systems—the immunity systems, the biologic amplication systems, and even the effector mechanisms—work, reflects again in almost every instance a constructive interplay

between the clinical study and the basic laboratory analyses. For example, we will talk a great deal in this symposium about the importance of the early components of the complement system because patients who are born lacking C1r, C1s, C2, C3 or C5 have had much extraordinary clinical difficulties as they try to get along without these single components of their crucial biologic amplification system, the complement cascade. This methodology, I am again espousing of paying close attention to the "experiments of nature" represented by those rare cases of human disease where a crucial component has been left out or is deranged,[22] has been the veritable wellspring of all of our new knowledge of the lymphoid system and the bodily defense and should surely be uppermost in the considerations of this workshop. Critical questions are regularly asked by the experiences with patients in the clinic. These questions, in turn, when properly focused, lead to development of model systems in the laboratory. The latter, when we use the powerful tools of modern scientific methodology, can be thoroughly analyzed. The information gained can in turn finally be established as a useful analysis only when it can be returned to the clinic as a new method for treatment or prevention of these same rare diseases. I surely hope and anticipate that our conference will expand this pragmatic element of our discourse. Indeed, as I look back over the history of immunology, I see that almost all of the real strides in this field have been made in the same way. The initial impulse so regularly derives from consideration of a patient's problem in the clinic. Thus, some of the most critically focused questions that have guided development of understanding and organization or function of the immunity systems have been derived from study of patients born with congenital, sometimes hereditary abnormalities of immunodevelopment and function. These highly focused questions have then been carried back to the basic laboratories for extensive hopefully definitive study to yield modern understanding of the development and function of the immunologic apparatuses. The crucial role of the biologic amplification systems and the effector mechanisms in the body economy is now being gained in exactly this same way just as has our understanding of the specific components of the immunity system. Basic science is most satisfying and most meaningful when it yields results of value to man. In our field where the to and fro interaction between clinic and laboratory is very prompt, satisfactions have always been immense and will continue to be immense as long as we insist on this most productive proximity of clinic and basic laboratory.

It should not be necessary here to reiterate, in detail, the concepts concerning differentiation and development of the 2 separate immunity systems that have developed over the last 15 years. We now know that cells from the yolk sac, travel in succession to fetal liver, and then to bone marrow and that they can develop under the influence of a central lymphoid organ, and through a process involving extensive proliferation and differentiation in that central organ, ultimately to be exported as cells in 1 of 2 separate populations of lymphocytes and lymphoid cells. To me, it is remarkable that already at the start of this conference we can precisely define and enumerate in quantitative terms in circulation and lymphoid organs the very elements of the 2 separate lymphoid systems. We have Boyse and Old's TL, Thy 1 or θ and LY series of allo antigens[23] in the mice as beautiful models to be used as our pattern. Further, there are the specific thymic antigens that in mice can be identified by immunization of xenogenic recipients. Such antigens are already acting as a prototype and are being developed for use in man. At least 100 antisera have already been prepared which seem to be able to identify the T lymphocytes immunologically. The B cells can be identified in chicken, mouse and man by their capacity both to synthesize and to secrete immunoglobulin molecules. In the bird, it is the bursa of Fabricius that seems to be responsible for differentiating this entire population of cells. Further, bursa cells can already be used completely to correct, over a long term, agammaglobulinemic chickens, that have been produced experimentally in several ways. However, not only can cells of the bursa achieve this correction, but after the bursa involutes, both bone marrow and the spleen contain cells that can produce similar long-term reconstitution.[24-27] Where is the bursa in man? That was one of our problems in the last conference, and it remains one of our central questions today. Location of the bursa of Fabricius or of the bursa 1 equivalent in man is still a subject that needs resolution. I still think Peyer patches and the lymphoepithelial-appendiceal tissue as exists in a rabbit is a very good bet as the basis of at least some of this development. Fetal liver and other gut-associated lymphoid tissues must also be considered and even bone marrow cannot be dismissed.

Our molecular understanding of immunity has brought us to the point of talking about domains of molecular configuration by which the function of antibodies is exercised and can be analyzed. I would like to urge at this conference that we begin to think of much larger immunologic domains. For example, we have the local antibody system that Hanson, Tomasi, Kunkel, Hong, South, Cebra and others have analyzed. This domain, as we will hear later in our workshop, also has a major component vis-a-vis cell-mediated immunity. Henney and Waldman[28] have taught us that in the pulmonary domain cellular as well as humoral immunities exist, and in my laboratories Jurg Muller has uncovered evidence for an enteroenteric system pertaining not only to humoral but also to cellular immunities. The Peyer patches with their highly specialized surface epithelium, which I have repeatedly emphasized, have a defining morphologic characteristic and are very similar to the surface epithelium of the bursal follicles, seem, from these and earlier studies by Hess,[29] to be especially geared to permit passage of particulate material. The new work of Bockman and Cooper[30] says that ultrastructurally the bursal surface epithelium is specialized like Peyer patches' epithelium to permit particles to cross the membrane. Further, Meuwissen and Van Alten[31] now tell us that they can stimulate the bursa to respond by injecting particulate antigen into the bursal cavity.

Can we think further of the skin as part of a peripheral domain? Why not think of a central or systemic immunologic domain? I think such considerations may be most useful and the consequences may have a powerful

influence on our manner of using our immunity systems in the future. Can it be that the B-cell development is also antigen driven in a special way? This we will consider in detail later in the conference.

The development of the full population of B lymphocytes in the circulation occurs so early in the development of the human and mouse fetus that one group of investigators like those led by Cooper[32] have proposed that up to an advanced stage, the development of the B-lymphocyte population is independent of antigenic stimulation. By contrast, plasma cells or secretory lymphocytes to their view do not develop until exposure to antigenic stimulation induces a terminal differentiation to antibody producing and secreting cells.

In our laboratories we look hard for things that do not fit. When these turn up, as they will in this workshop, we must pay close attention. I will mention one such finding in passing and urge our serious reflection on it. Max Cooper and his co-workers have taught us that the development of B lymphocytes during the embryology of the human fetus occurs long before appearance and full development of Ig-secreting lymphocytes or plasma cells. The views of the Cooper school in this regard are exciting. But are they correct? Let us pay especially close attention to the pig which is separated from all antigens in such an extraordinary way by the 6 layered epithelial and vascular placenta. The pig does not seem to show this same B lymphocyte development. Indeed, B lymphocytes do not appear, or if they do appear, they are present in very small numbers possessing only surface IgG until the piglet is exposed to antigen in extrauterine life.[33-38] Perhaps we should begin to think here in quantitative terms with respect to an antigen-based drive for development of B cells. Perhaps a different amount of antigen controls development to B lymphocytes on the one hand and the terminal differentiation to secretory B cells on the other. Certainly, once the pig is exposed to antigen, it quickly develops both the B-cell population and Ig-secreting cells. We need to think seriously about this issue, to analyze it further in our discussions, and to do the experiments necessary to reject or encompass the findings and thus deal with the postulates of the Cooper school. It is important to remember that a really useful scientific hypothesis is one that can be disproved with a single well-designed experiment.[39]

In man we can already specifically recognize and quantify both the T- and B-cell populations, and we have the prospect of reproducible quantification of the faults that exist in patients with immunodeficiencies, that are either genetic or acquired. We can monitor serially each system in morphologic and functional terms.

The whole business of cooperation between B and T cells is something that needs much more attention. In spite of the fact that I protested, it is already clear in some experimental systems that IgG antibody synthesis, particularly in secondary responses, does involve in certain species an extraordinary cooperation between T and B cells. How obligatory is this cooperative function remains a pregnant question.

The cooperative enterprise between T and B cells has had a stormy history. It was first argued, you remember, that the thymus-derived cells function as the recognition units and in some way transfer to the thymus-independent or bone marrow-derived cells, specific information which then serves to use the marrow-derived cells as factories to do the work of Ig synthesis as directed by informational machinery of the T lymphocytes. But no information was transferred, and it became clear that T lymphocytes acted in some other way to facilitate B-cell function. Then Mitchison[40] and Bretcher and Cohn[41] conjured up the idea that T cells and B cells interact directly with antigen, sitting somewhere in the middle to give an essential two-impulse stimulation to the B cells, and thus to induce them to form antibody. This concept now seems quite unacceptable in light of more recent studies led by the finding of Katz *et al*[42] that a graft-vs-host reaction of T lymphocytes could substitute for the T-cell influence. Now it seems that T cells produce *something* which directly or indirectly influences B cells and makes them more capable of responding to antigens. We badly need a *molecular definition* of this cellular interaction and we need to know how important it is for man. I predict that the clearest way of identifying its relevance will be to find an "experiment of nature" in which on genetic grounds the capacity for such cooperative function has been left out. We would then quickly see its importance and meaning in the body economy.

Remember that, before the patients with the Bruton-type or X-linked infantile agammaglobulinemia became available, authorities were arguing as to whether antibodies had anything to do with recovery from infection. Now of course, we know patients with this form of immune defect, where there is apparent integrity of the T cells and T-cell function and a veritable absence of the B-cell system and antibody responses, are in great jeopardy from infections with high-grade encapsulated bacterial pathogens. They cannot survive without the system unless very effective antibodies are available. The age of these patients is just exactly that of penicillin. Not because penicillin or any other antibiotics have caused the disease, as has been suggested, but because the disease as recurrent infection with high-grade encapsulated bacterial pathogens could not exist until antibiotics were generally available to rescue these patients from their individual bouts of infection with these encapsulated bacterial pathogens. It is thus clear that the B-cell system per se is vital to recovery from infection and thus survival. The T-cell system is also vital to life because in the presence of an apparently adequate B-cell immunity system, the patients with DiGeorge syndrome, born without a thymus and with a grossly deficient or absent T-cell system, do not survive. This does not gainsay that in some patients with DiGeorge syndrome, the T-cell deficiency is incomplete and that such patients can survive for a prolonged period.[43] Nonetheless, absence of the T-cell system is a highly lethal defect. These "experiments of nature", as examples of many others, have helped us comprehend the crucial role of each of the 2 specific immunologic functions. Yet we still do not understand the *molecular* basis of T-cell function. This is particularly apparent when we use our "experiments of nature" to make available for study, systems that contain a minimum of noise such as exists with the blood and lymph node cells from agammaglobulinemic children. Another

system relatively free of noise is the agammaglobulinemic chicken model prepared by in ovo bursectomy. We can ill afford the confusion contained in many experiments that produce false information from poorly chosen models or systems where the noise level is so great as to have led to extravagant conclusions about the molecular basis of T-cell immunity. As examples are the studies by Marchalonis and others who attribute the specificity of T-cell immunity to immunoglobulins. Far too many experiments are being done without using the full benefit of the capacity of appropriate clinical and experimental models to clear out much of the confusing noise, and to permit analysis of this vital but difficult question in molecular immunobiology under the best possible circumstances. Our own concepts of IgX, although they must be tested, have not yet been confirmed and seem at the moment a long distance from reality.

Similarly, patients lacking specific components of each of the biologic amplification systems and each of the key steps involved in the operation of the effector mechanism, either already have been or will be discovered. They will each have the same kind of vital importance to the advancing of knowledge as do the patients with the primary immunodeficiency diseases. The nature of the disease presented by each of these patients, the specific nature of their vulnerability to exogenous or endogenous pathogens, has helped or will in the future help to reveal the critical role of the cells, molecules or functions being considered in the body economy. Here again the contributions of these infrequent patients has been and will continue to be most revealing in another way. As knowledge improves and as we better and better understand the bodily defenses, we will become more and more capable of making reasonable efforts to correct the abnormalities of structure and function of the systems of defense. As an example, almost immediately after the last conference, Cleveland[44] was able to transplant a thymus to a patient with DiGeorge syndrome apparently to correct the immunologic defect of cellular immunity so characteristic of patients born without a thymus. Confirmation came quickly from August *et al*[45] and then from Gatti *et al*[46] and Biggar *et al.*[47,48] To really be sure about this correction, however, it has been essential to go again to the model systems in animals where no T cells exist, and to show in these models that thymic transplantation, even just little wet membranes of embryonic thymus like Biggar *et al* have used,[49] will correct entirely the T-cell defect. No, or very little, T-cell immunity develops in the absence of the thymic influence. Thus, since the last conference it has been established for man and the mouse that the epithelial or embryonic thymus can correct the T-cell deficit whether that defect is genetic or iatrogenic. These are dramatic examples of cellular engineering. More than that, Cleveland's achievement can now be extended to other T-cell deficiencies, as we will be told by Kirkpatrick *et al*[50] of the NIH and as Aiuti, Gatti and co-workers from Rome have so ably demonstrated.[51] It is exciting to realize that we now can monitor precisely such corrections, because in man we can precisely quantify numbers of T lymphocytes and B lymphocytes and can quantify their responses and functions so much better than we could at our last international workshop in 1967.

In the recent work of Bach,[52] Goldstein and White,[53] and of Boyse and Komuro,[54] we have the possible anticipation of being able to achieve such cellular engineering at a molecular level.

Our bone marrow transplants have corrected severe combined immunodeficiency (SCID) since the last meeting. These are surely most dramatic achievements. They can be viewed as essentially creating life for these patients with SCID by providing both of the 2 immunity systems in children born without adequate amounts of either system. I hope at this meeting we will be able precisely to enumerate the successes in correcting SCID by marrow transplantation. In my own crude counting, I know there are somewhere around 25 such successful resolutions of these rare but devastating diseases.

At this meeting, we see by the abstracts that Dr. Day will tell us how she has already observed correction of inborn errors of the complement system by another form of cellular engineering, namely by renal transplantation. Thus, intensive studies of patients with primary and secondary immunodeficiencies will, I am sure, continue to demand our attention as the source of critical questions. Thus, the study of these patients will continue to provide relevance to our laboratory inquiry. Finally, these patients will test our developing knowledge and our capacity to predict and to control maximally those events that relate to the development and function of the immunity system. Similarly, anomalies in which the complement system is perturbed or incomplete will continue to guide our understanding of the role of this extraordinary cascade in the body economy. I was astonished to see from the abstracts that Dr. Alper still maintains that the specific component defects of the complement system, except for defects of C3, ordinarily do not cause disease. This issue should be settled in our conference. Dr. Alper's view conflicts with the findings of Dr. Day and Drs. Agnello and Kunkel, as well as with the findings of Miller and Shin and me. Hopefully we can present evidence that will convince him at long last of the importance of each of the complement components in the body economy.

I remember well when the Boston group, some 8 or more years ago described their immunologists and families of immunologists who they found to lack the second component of complement. They had recognized that these people were quite healthy, and drew the conclusion that this component and maybe the entire complement cascade was irrelevant. I could not wait to get the microphone to disagree because the earliest phylogenetic studies carried out by Gewurz, Finstad and me[55] contained evidence indicating that the complement cascade had been maintained in essentially the same form for between 300 and 400 million years. Such a cascade of interacting proteins, capable of acting as a biologic amplification system for immunity, could not have been maintained without having had major survival advantage as a system. Defects of many of the components of this system, even defects of subcomponents of the first component of the system, can now be shown to be associated with and to underly highly lethal diseases. The absence of any component reveals disease that will reflect the survival advantage of both the component and the system of interacting components. We can expect to find dis-

eases associated with defective development and function of cells and processes that are involved in inflammation, phagocytosis, blood coagulation, smooth muscle contraction and vascular reactivity. These diseases, like those associated with immunodeficiency and defects of the complement system, will teach us much about the real role of these individual effector mechanisms in the body economy.

Because of the broad generalizations that can be derived from analyses of the relatively infrequent patients with immunodeficiency diseases, the importance of intensive study of these inborn abnormalities can be expected to contribute far more than just understanding of their own disease. Study of the disease will also achieve in turn effective treatment for their illnesses. Studies of patients with primary and secondary immunodeficiencies, have helped, are helping and will continue to help us understand the normal immunity system. They will also help us to understand the perturbations and functions of these systems that underly or are an important part of many of man's most frequent and most distressing diseases. The philosophy that underlies this concept is most beautifully expressed in the letter that many of you remember, which was written by William Harvey in the twilight of his life[22]:

> "Nature is nowhere accustomed more openly to display her secret mysteries than in cases where she shows traces of her workings apart from the beaten path, nor is there any better way to advance the proper practice of medicine than to give our minds to the discovery of the usual law of nature, by careful investigation of cases of rarer forms of disease, where it has been found in almost all things that what they contain that is useful or applicable is hardly perceived unless we are deprived of them or they become deranged in some way."

Thus, although the primary immunodeficiency diseases are infrequent, the knowledge to be gained from their study can be extended to address many of the problems encountered in diseases of man that represent the most common medical problems.

In launching our conference, it is well to reflect on a few examples of situations where major perturbations of the immunity functions exist, that can and will be understood through knowledge obtained in an important part from study of patients with the rare primary immunodeficiency diseases.

1) Significant and even severe depressions of immune function are encountered during the aging process in man and all mammals thus far studied. Nothing is more consistent throughout phylogeny than the apparent programmed involution of the thymus and thymus-dependent system.

2) Many malignancies, particularly those involving cells of the immunoapparatus, but also many others including carcinomas and sarcomas, in their widely disseminated state, are accompanied by depression of cell-mediated and/or humoral immunities. They are accompanied by profound disturbances in immunologic balance.

3) Oncogenic viruses and chemical carcinogens very frequently have immunosuppressive properties.

4) Many common virus infections, which have long been known clinically to open the door to serious bacterial infections or even to more serious virus or fungus diseases, can depress immunity functions, sometimes most profoundly.

5) Certain bacterial infections like leprosy and tuberculosis can be associated either with stimulation or depression of immunity potential and immunity function.

6) Similarly, fungus or protozoan infection can depress immunity. For example, recent work of Sven Gaard and his associates at the Karolinska, has shown a profound immunodepression to be associated with experimental and clinical toxoplasmosis.

7) Patients who develop autoimmune diseases often do so because there exist abnormalities or deficiencies of the normal immunity function. It has become even more clear, for example, that autoimmune diseases are often consequent to the invasion or extension of infections when the immunity system is perturbed, depressed or disbalanced. Instead of being due to forbidden clones, as was proposed by Burnet[56] and Fudenberg,[57] it seems to us that autoimmunity is regularly due to forbidden antigens like those associated with infection that are often handled most inadequately by a malfunctioning or disbalanced immunologic system.

8) Toxic compounds like alcoholic beverages, cigarette smoke or environmental pollutants can adversely influence important immunity functions. For example, the functions of alveolar macrophages are profoundly depressed by intake of excessive amounts of alcohol and T-cell defects are produced.

9) Chronic severe malnutrition can profoundly depress humoral and cellular immunities and, depending on the acuteness or chronicity or upon the degree of nutritional deprivation, can unbalance a system that must function in delicate balance to be most effective in the bodily defense.

10) Many of the most powerful anticancer drugs or other agents that we use in modern medicine can act as immunosuppressants or can interfere with biologic amplification or effector mechanisms. Even extensive surgery can produce profound perturbations of the immune system or can facilitate immunodeviation.

11) Organs, tissue and cellular transplantation, now widely applied, in the form of halfway treatment measures in clinical medicine require, at present, carefully controlled depression of immunologic function. The control, unfortunately, at this point is often not quite precise.

Thus, dissection and analysis of the immunologic system, to which study of the immunodeficiency patients has contributed so much, is contributing and will continue to contribute, is necessary to help us to develop the capacity to modify, modulate and even engineer effectively the prevention, or treatment of adverse manifestations of some of the most common and most devastating diseases and disorders that man faces today. I believe that the most effective way of continuing to approach the difficult questions and problems of clinical immunobiology is to continue to carry out intensive inquiry into the nature of the primary immunodeficiency diseases of man. The basic analyses of these diseases will surely enhance our efforts to correct these primary immunodeficiency diseases.

Efforts to correct the defects in children with primary immunodeficiency diseases was seriously criticized by F. M. Burnet at the First International Immunology Congress in 1970.[58] Nonethe-

less, I cannot take his criticism seriously. His suggestion that we do not try to correct the deficiencies in these rare patients I find absolutely unacceptable as do almost all physicians with whom I have discussed this issue. By attempting to correct the deficiencies in patients with primary immunodeficiency disease, we are testing our developing knowledge and by so doing we are developing the important discourse of clinical immunobiology. We are developing our capacity to use cellular engineering and even molecular engineering. We must continue to test these developing strengths until they can be applied to the management of the major remaining serious diseases of man. To propose that by treating these rare genetically-determined diseases we will seriously alter equilibrium gene frequencies or put intolerable, or even a significant, burden on civilization seems to me to ignore facts already handled well by others concerning the influences of modern medicine on genetic equilibria.[59]

I would urge all of you to listen, to reflect upon, and then to reject such unfortunate criticisms of our endeavors. You must continue your most important studies. You must present your achievements with pride. You must be prepared to attack one another's findings without restraint because it is only in the face of the most stringent criticism that we can expect to make the kind of solid advances that are needed for the further growth and development of this important discourse. Let the attacks be on one another's works and not on one another. Science must be built of solid and durable material, but effective creative productive scientists are precious and sometimes rather delicate commodities.

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