Immunity
Forum

A Decade of Vaccines: Integrating Immunology and Vaccinology for Rational Vaccine Design

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Vaccination stands as one of the most successful public health measures of the last century. New approaches will be needed, however, to develop highly effective vaccines to prevent tuberculosis, HIV-AIDS, and malaria and to eradicate polio. Current advances in immunology and technology have set the stage for rational vaccine design to begin a "Decade of Vaccines."

Introduction: A Brief History of Vaccines

The notion of protective immunity can be traced back to the observation in the fifth century BCE that individuals who had recovered from disease during the Plague of Athens were protected from subsequent attacks. However, the birth of the science of immunology is most readily attributed to the demonstration by Jenner at the end of the 18th century CE that individuals intentionally inoculated with material from cowpox-infected cattle were protected from smallpox. This demonstration predated evidence for the microbial (i.e., germ) origin of infectious diseases obtained by Koch and Pasteur. It also predated the elucidation of the immunological factors underlying this protective effect by von Behring and many others. These "immunologists" went on to develop this field as a discipline and to illuminate the crucial role of immunity and inflammation in infectious diseases and in many other aspects of human physiology (Allen et al., 1999).

Over the years, the fields of immunology and clinical vaccinology diverged: immunology became progressively focused on model systems that allowed its intricacies to be probed in cellular and molecular detail, whereas vaccinology addressed more practical problems, focusing on humans and other species for which vaccines were intended. Absent a knowledge-based toolkit by which to reliably induce protective immunity to the pathogens of interest, vaccinology has been left to rely almost solely on empirical, trial-and-error approaches not so different from Jenner's, approaches that seek to mimic the processes of natural infection while reducing the untoward effects to an acceptable level.

Nonetheless, vaccinology has been a stunning success, with vaccination being one of the greatest public health measures of the past century, and arguably the most cost effective of all (Centers for Disease Control and Prevention, 1999). The eradication of smallpox in 1977 is a landmark achievement. The potential for eradication of polio is at hand, although both public health and immunobiological challenges remain (Serazin et al., 2010). The development of rotavirus vaccines offers the promise of saving the lives of the more than 500,000 young children worldwide who die from diarrheal illness caused by this virus each year (Madhi et al., 2010; Richardson et al., 2010). *Haemophilus influenzae* type b, pneumococcal, and meningococcal polysaccharide-protein conjugate vaccines have been a major success in the developed world countries where they are in common use, reducing and in some cases nearly eliminating pneumonia, sepsis, and meningitis due to these pathogens. And if the benefits of these conjugate vaccines can be extended to children in other parts of the world, more than one million childhood deaths could be prevented (http://www.who.int/immunization_monitoring/burden/en/).

These conjugate vaccines have achieved such great success because they convert the antibody responses to the target polysaccharide from T cell-independent to T cell-dependent responses. As a result, the vaccines are vastly more immunogenic in the young children at greatest risk than is the infection with the pathogen itself. This Lasker Award-winning achievement stands alone among the achievements to date in vaccinology as the only clear example of rational vaccine design driven by and dependent on knowledge derived from the fundamental immunological observations—made by Landsteiner, Avery, and Goebel—decades earlier (http://www.laskerfoundation.org/awards/1996clinical.htm).

Although there has been a welcome and substantial increase in vaccine discovery and development efforts within industry and in the public sector in the recent past, this increase has not been matched by a comparable increase in the novelty of vaccine concepts or in the predictability of the process. Surely, vaccine discovery and development can become more rational, and they must do so to achieve the progress envisioned by Bill and Melinda Gates and expressed as a challenge at the 2010 World Economic Forum. Their challenge was to make this the "Decade of Vaccines," a decade in which eight million children would be saved from deaths potentially preventable by vaccines. To succeed, recent gains in our understanding of basic immunology, microbial pathogenesis, and immune evasion, together with technological innovations in a variety of other fields, must be applied to create, test, and refine candidate vaccines; to study the response to vaccines in humans in vivo in a more holistic, expeditious, and iterative manner; and to refine animal models so they are more informative and predictive of human vaccine responses.

The State of the Art of Vaccine Immunology

The series of Reviews in the current issue of *Immunity* provide a complementary, contemporary perspective on the issues described above. These articles discuss

the immunological basis of vaccine science, including the determinants of vaccine-induced immunity; tools for the induction of effective immunity; conceptual frameworks to identify vaccine targets and seek causal correlates of protection; and progress toward effective vaccines for tuberculosis, HIV-AIDS, and malaria. Below we highlight some key elements from each review article.

Vaccine-Induced Immunity: Molecular, Cellular, and Anatomical Determinants

Three articles in this issue together cover the molecular, cellular, and anatomical determinants of vaccine-induced immunity. Vaccination induces immunological memory that protects against subsequent natural infection by a pathogen. Sallusto et al. (2010) describe a process for vaccine development—that they term analytic vaccinology— based on analyzing memory B cells and memory T cells to understand the molecular basis by which they can provide protection against infection with particular pathogens. The observed effects can then be refined and enhanced as part of rational vaccine design, seeking not only to optimize the magnitude but the duration and functional qualities of memory T cell and B cell/plasma cell responses. Sallusto et al. cite several studies, for instance, that use newly developed technologies to comprehensively characterize the human antibody response to infection. Despite the diversity of HIV-1 and influenza viruses, these studies identified broadly neutralizing antibodies for each virus, thereby identifying conserved epitopes which can guide new vaccine development.

Diverse pathogens invade at mucosal surfaces, and a localized mucosal immune response is required to protect against such invaders as HIV and *M. tuberculosis*. Mucosal vaccines—those administered orally or by inhalation— can induce the production of antibodies that inhibit the earliest steps in infection, including pathogen attachment, but few mucosal vaccines have been successfully developed to treat human infections. Chen and Cerutti (2010) attribute this lack in part to an incomplete understanding of the finely tuned nature of mucosal immunity, which evolved to detect pathogens while balancing tolerance to the vast community of microbes inhabiting mucosal surfaces. A major challenge is to induce sustained mucosal immunity while not perturbing this balance either toward overstimulation and consequent inflammation or toward counterproductive tolerance. In addition, challenges in developing a broadly effective vaccine arise because of mucosal physiology specific to the elderly and to women. Nevertheless, as Chen and Cerutti (2010) further note, the complex regulation of mucosal immunity also provides opportunities, such as the potential to enhance immune response by dietary supplementation with vitamin A, perhaps particularly relevant for populations with inherent nutritional deficiencies.

Dendritic cells, once activated by foreign antigens via the pattern recognition receptors of innate immunity, initiate an adaptive immune response to these antigens. As discussed by Palucka et al. (2010), the type of immune response initiated depends on the type of dendritic cell and also on the particular innate immune signals received. These properties make dendritic cells attractive potential targets when designing vaccines to produce a specific immune response. Palucka et al. (2010) note, for instance, that plasmacytoid dendritic cells found in blood have a number of features that could make them good targets for new antiviral vaccines. Similarly, on the basis of evidence that Langerhans cells, a dendritic cell subset found in the skin, are functionally specialized to activate cellular immunity, Palucka et al. (2010) propose that these cells may be good targets of vaccines designed to prevent chronic diseases, including tuberculosis, HIV/AIDS, and malaria. A key remaining challenge in such vaccine design is determining the best mechanism to target the dendritic cell subset.

Tools for the Induction of Effective Immunity

When there is a need to increase the immune response to a vaccine or to alter the types of induced immunity, there are powerful tools at hand. These tools are particularly relevant for targeting pathogens for which natural infection does not induce effective immunity. Adjuvants, vaccine components that enhance immunogenicity, are one such tool. Complete Freund's adjuvant, developed empirically and long used in experimental systems, consists of heat-killed mycobacteria in a water-in-oil emulsion formulation, with both the source of antigens and the formulation contributing to activity. As discussed by Coffman et al. (2010), there is a current emphasis on rationally designing adjuvants on the basis of known correlates of immune protection, rather than the empirical approach used historically. Most adjuvants are thought to work primarily by stimulating innate immunity, and they are most effective when used in combination to stimulate multiple immune pathways, as would be the case during natural infection or with live, attenuated vaccines (Coffman et al., 2010). An excellent example highlighted by Coffman et al. (2010) is the RTS,S malaria vaccine, which conferred protection that was dependent on the combination of adjuvants. Coffman et al. (2010) conclude their Review by noting that adjuvant research leading to clinical trials, even though highly directed to prevent disease, may ultimately yield a wealth of data on the immune responses of healthy humans.

Another tool that can be used when live, attenuated vaccines are not feasible is to deliver pathogen antigens by vectors. Vaccine vectors include viruses, bacteria, DNA, and RNA. As discussed by Liu (2010), vectored vaccines can be exquisitely tailored in terms both of the cell types and cellular compartments targeted and in terms of how the antigens are delivered. One striking example highlighted by Liu (2010) is a successful veterinary rabies vaccine, incorporated into food bait, which used an altered version of the human smallpox vaccine (modified vaccinia Ankara) as a vector to deliver rabies virus antigen to multiple wild animal species. The immune response to the vector itself must be taken into account during vaccine development, and it was a source of concern in the STEP trial of an HIV vaccine employing an adenovirus vector. On a positive note, Liu (2010) emphasizes that the efficacy of vectored vaccines can be enhanced by using two different vectors—or two different types of vaccines—in series in prime-boost immunization, and mixed modality prime-boost immunization trials with HIV vaccines are currently underway.

Conceptual Frameworks

One common theme in this issue's vaccine review series is the importance

of taking a holistic view of the immune system. Two conceptual frameworks that emphasize taking the broadest possible perspective are reverse vaccinology (Sette and Rappuoli, 2010) and "systems vaccinology" (Pulendran et al., 2010). Reverse vaccinology begins with bioinformatic analysis of a pathogen genome to comprehensively identify antigens in silico. Candidate antigens are then progressively eliminated by experimental tests until candidates for vaccine trials remain. This sequence is a reversal of the usual work flow in which extensive analysis requiring culturing the organism comes first and bioinformatic analysis later. In its first usage, reverse vaccinology quickly yielded a type b meningococcal vaccine candidate, which had previously seemed out of reach. In a similar "reverse" strategy used in a recent study highlighted by Sette and Rappuoli (2010), in silico prediction of MHC-binding vaccinia virus peptides yielded a comprehensive list of the epitopes responsible for the murine T cell response to the virus. Sette and Rappuoli (2010) note that such strategies for generation of unbiased and comprehensive antigenic maps of pathogens are likely to be widely applied in the coming years.

Systems vaccinology—a systems biology approach to vaccinology—attempts to capture the network of relationships that integrate the parts of the immune system, from molecules to cells to tissues, and use it to predict the functioning of the system as a whole as it applies to vaccine science. In one example discussed by Pulendran et al. (2010), human gene expression signatures were identified that could predict the $CD8^+$ T cell response induced by the highly successful yellow fever vaccine, and these signatures implicated the integrated stress response in vaccine-induced immunity. Extending such studies to other vaccines could yield sets of gene expression signatures, each predictive of a different facet of vaccine immunogenicity. Integrated into a "vaccine chip," these signatures could be used both to guide the design of new vaccines and to identify vaccinated individuals with suboptimal responses (Pulendran et al., 2010). Pulendran et al. point out that a systems biology approach could have many additional uses. The approach could be used to develop co-correlates of protective immunity that might reflect vaccine efficacy better than the single variables commonly used and might be applied when vaccine efficacy is determined by a balance between elements of humoral and cellular immunity, for instance. More generally, this is a powerful type of approach for revealing new biology and could be used to systematically determine the detailed mechanism of action of adjuvants so that they can be used most effectively.

Although systems-level analysis in biology is not a replacement for more focused efforts, both Pulendran et al. (2010) as well as Germain (2010) discuss some of the research programs being created to facilitate such analysis and capitalize on the latest technologies. Akin to the multiple efforts underlying the human genome project, these programs will encompass a consortium of human immune profiling centers, academic institutes, and linked government laboratories. The common goal is to characterize the human immune system in health and when perturbed by infection, vaccination, or genetic disease. These research programs will help bridge basic research on vaccines with ongoing clinical trials, ensuring not only that basic research guides clinical trial design but also that information gained from clinical trials is used to drive basic research (Pulendran et al., 2010). The opportunities for new discovery added by this cycling of information will in turn help ensure that during vaccine design what should be measured always trumps what can be measured (Germain, 2010).

Vaccines for Tuberculosis, HIV-AIDS, and Malaria

For three diseases that are critical global health threats—tuberculosis, HIV-AIDS, and malaria—there are no highly effective vaccines. Three perspective articles in the current issue describe the vaccine research underway directed against these diseases. Kaufmann (2010) reviews the vaccination strategies directed against tuberculosis. He notes that the current live attenuated BCG vaccine protects against severe disease in infants, but is ineffective against adult pulmonary disease. Despite the BCG vaccine being given four billion times since its first use 90 years ago, its mechanism of protective immunity is unclear. Nevertheless, of the 11 candidate vaccines now in clinical trials, two are recombinant forms of BCG and seven are subunit booster vaccines to follow priming with BCG; the remaining two candidates do not contain live bacteria and are not meant to be administered with BCG, given that they are targeted to individuals coinfected with *M. tuberculosis* and HIV (Kaufmann, 2010). All these candidate vaccines aim to delay active disease, not to prevent or eliminate infection, which are goals for the future. Echoing Pulendran et al. (2010), Kaufmann (2010) concludes that achieving these two future goals is likely to require effective cycling of information between basic research and clinical trials.

McElrath and Haynes (2010) review the vaccination strategies directed against HIV-1. They note the many aspects of HIV-1 biology that complicate development of a vaccine, including the fact that HIV-1 evolution within its hosts has created a worldwide level of viral diversity perhaps beyond the reach of a single vaccine candidate. In addition, key data that drive rational vaccine development remain unknown for HIV-1, such as correlates of protective immunity and how predicted correlates might best be elicited. Despite these challenges, the recently published results of the RV144 vaccine trial conducted in Thailand demonstrated for the first time that a vaccine regimen could reduce HIV-1 infection rates, in this case by 31%. The RV144 trial used a recombinant canarypox vector expressing three HIV-1 proteins as a prime and two different recombinant HIV-1 gp120 envelope glycoproteins with alum adjuvant as a boost. The observed reduction in infection, albeit modest, is certainly grounds for optimism, and determining the correlates of protective immunity in this trial is one avenue forward (McElrath and Haynes, 2010).

Good and Doolan (2010) review the vaccination strategies directed against malaria. As is the case for developing vaccines targeting HIV, a challenge in targeting the malaria parasite is its diversity, not only because of the different *Plasmodium* species that infect humans, but also because of genetic mechanisms inherent in *Plasmodium* that generate surface antigen diversity. This challenge notwithstanding, there has been recent success in malaria vaccine efforts. The RTS,S vaccine, consisting of a fusion protein

antigen—the *Plasmodium* circumsporozoite protein fused to the hepatitis B surface antigen—combined with an adjuvant mixture, reduced infection in African children by 30%–50%. For the vaccine version currently in phase 3 clinical trials, the adjuvants are monophosphoryl lipid A and the saponin QS-21 in a liposomal formulation. Good and Doolan (2010) suggest that in parallel to developing vaccines based on immunodominant antigens, such as the circumsporozoite protein, it is also important to move forward with research aimed at inducing a broader immune response, such as by using live attenuated parasites.

Concluding Remarks

This series of *Immunity* Reviews frames a number of the key issues in vaccine science and identifies opportunities for productively integrating immunology and vaccinology. A closer working partnership between these fields and with colleagues in information, engineering, bioengineering, and biomaterials science is needed. We propose that through such partnerships lies the path to accelerate the development of vaccines for major global disease threats. Knowledge gained through these partnerships will help to select which are the best candidates to enter human vaccine trials, thereby increasing the efficiency of these trials, and ultimately will promote the creation of vaccines that are less costly and more practical for people in resource limited settings where the burden of disease is greatest.

REFERENCES

Allen, P.M., Murphy, K.M., Schreiber, R.D., and Unanue, R.D. (1999). Immunity *11*, 649–651.

Centers for Disease Control and Prevention. (1999). MMWR Morb. Mortal. Wkly. Rep. *48*, 243–248.

Chen, K., and Cerutti, A. (2010). Immunity *33*, this issue, 479–491.

Coffman, R.L., Sher, A., and Seder, R.A. (2010). Immunity *33*, this issue, 492–503.

Germain, R.N. (2010). Immunity *33*, this issue, 441–450.

Good, M.F., and Doolan, D.L. (2010). Immunity *33*, this issue, 555–566.

Kaufmann, S. (2010). Immunity *33*, this issue, 567–577.

Liu, M.A. (2010). Immunity *33*, this issue, 504–515.

Madhi, S.A., Cunliffe, N.A., Steele, D., Witte, D., Kirsten, M., Louw, C., Ngwira, B., Victor, J.C., Gillard, P.H., Cheuvart, B.B., et al. (2010). N. Engl. J. Med. *362*, 289–298.

McElrath, M.J., and Haynes, B.F. (2010). Immunity *33*, this issue, 542–554.

Palucka, K., Banchereau, J., and Mellman, I. (2010). Immunity *33*, this issue, 464–478.

Pulendran, B., Li, S., and Nakaya, H.I. (2010). Immunity *33*, this issue, 516–529.

Richardson, V., Hernandez-Pichardo, J., Quintanar-Solares, M., Esparza-Aguilar, M., Johnson, B., Gomez-Altamirano, C.M., Parashar, U., and Patel, M. (2010). N. Engl. J. Med. *362*, 299–305.

Sallusto, F., Lanzavecchia, A., Araki, K., and Ahmed, R. (2010). Immunity *33*, this issue, 451–463.

Serazin, A.C., Shackelton, L.A., Wilson, C., and Bhan, M.K. (2010). Nat. Immunol. *11*, 551–555.

Sette, A., and Rappuoli, R. (2010). Immunity *33*, this issue, 530–541.