Sometimes simple things are hard to handle. This is true of infectious diarrhea, which remains one of the leading causes of death in children worldwide and a major factor in long-term morbidity. Some gut infections rapidly become systemic, with deadly effects even in adults, as evidenced by the outbreak of illness associated with Shiga-toxin–producing Escherichia coli in Europe this summer. Vaccines have long been sought to protect the intestine from such pathogens and their toxins. However, the search has proved difficult because the conventional route of immunization — by injection through the skin — usually generates systemic immunity but provides little or no protection at mucosal surfaces, whereas mucosal immunizations target immune responses to mucosal tissues but are often hampered by insufficient potency, poor safety, or both. Thus, a new study reported by Hammerschmidt and colleagues warrants attention.

Effective immunization requires the processing and presentation of antigen by dendritic cells in juxtaposition to naive T cells. This presentation occurs only where the two cells meet in secondary lymphoid organs, such as lymph nodes draining the skin or organized lymphoid aggregates that collect antigens from mucosal surfaces such as those in the intestine (subepithelial Peyer’s patches and mesenteric lymph nodes), lung, and other sites that are open to the environment. In lymph nodes, dendritic cells migrating from the periphery, loaded with exogenous antigens and activated by “danger signals,” identify naïve T cells that are uniquely capable of recognizing the antigen and induce them to proliferate and to differentiate into one of several effector T-cell phenotypes. Dendritic cells also provide directions to newly minted effector T cells so that they can migrate, establish local immunity, and fight infection where required.

We now know how naïve T cells are imprinted by dendritic cells to target migration into the small intestine. Unlike dendritic cells in other lymphoid tissues, those in Peyer’s patches and mesenteric lymph nodes possess enzymes that convert dietary vitamin A into all-trans retinoic acid. Thus, the presence of all-trans retinoic acid is a characteristic feature of the lymphoid environment of the intestine (Fig. 1B). All-trans retinoic acid that is present during antigen presentation induces B cells and T cells to express on their surfaces the $\alpha_4\beta_7$ integrin that binds mucosal addressin-cell adhesion molecule 1 (MAdCAM-1) — an adhesion receptor specific to mucosal microvascular endothelial cells — and the chemokine receptor CCR9 that senses CCL25, a chemokine that is constitutively expressed in the small intestine. When the activated lymphocytes return from the mucosal lymphoid tissues into the blood, these two trafficking molecules (termed “homing receptors”) enable effector cells to migrate to the intestinal mucosa (Fig. 1). If all-trans retinoic acid is not present, however, such as when dendritic cells and naïve T cells meet in lymph nodes draining the skin, the T cells are induced to express other homing receptors that target the T cell back to the skin (Fig. 1C). The resulting effector cells can accumulate in the skin but travel poorly to the gut, which is why percutaneous vaccination often generates inadequate mucosal protection.

Although it is seemingly a simple proposition, immunization through the gut has been profoundly problematic because the intestine is organized to promote immune tolerance, not immunity toward most orally administered antigens; this is why people are usually not allergic to food. To get around this problem, Hammerschmidt et al. reasoned that immunizations applied by subcutaneous injection might induce intestinal lymphocyte homing (and thus intestinal immunity) if the injection also contained all-trans retinoic acid. This is exactly what they found. They immunized mice with chicken-egg
Subcutaneous injection of chicken-egg ovalbumin

Dendritic cell

Skin-draining lymph node

Skin-homing effector memory T cell enters circulation and returns to the skin

Antigen-presenting cell

T cell

All-trans retinoic acid

Skin-draining lymph node

Subcutaneous injection of chicken-egg ovalbumin and all-trans retinoic acid

Skin

Skin-draining lymph node

Gut-homing T cell returns to the intestinal mucosa through circulation

Antigen-presenting cell

T cell

All-trans retinoic acid

Intestinal mucosa

Small intestine

Oral ovalbumin

Gut-homing T cell returns to the intestinal mucosa through circulation

Peyer's patch and mesenteric lymph node

α4β7

CCR9

Peyer's patch

Subcutaneous injection of chicken-egg ovalbumin

Oral ovalbumin

Intestinal mucosa

Small intestine

Skin-draining lymph node

Skin

Oral ovalbumin

Small intestine

Intestinal mucosa

Skin-draining lymph node

Skin
ovalbumin applied subcutaneously with or without all-trans retinoic acid or by oral gavage of ovalbumin alone, administered directly to the gut mucosa (Fig. 1). To monitor trafficking of antigen-specific lymphocytes, the mice were infused with fluorescently tagged naive T cells. All-trans retinoic acid applied subcutaneously, together with ovalbumin, caused the transferred T cells to express the homing receptors for the intestine and to migrate there and function in numbers similar to those induced by oral immunization with ovalbumin alone, albeit at a dose 1000 times that of the subcutaneous injection. Remarkably, subcutaneous immunizations against cholera toxin and salmonella, when applied with all-trans retinoic acid, induced considerable protection against “disease” as modeled in the mouse. In short, the skin-draining lymph node was converted from a site specifying dermal immune defense to a site specifying intestinal (mucosal) immunity.

This is an important discovery, coincidently shared in part by Tan et al. Both groups have elegantly applied knowledge of how all-trans retinoic acid operates in antigen presentation to demonstrate a strategy for clinical application. This approach is attractive because immune responses in skin-draining lymph nodes are better defined and more vigorous than those of mucosal lymphoid tissues. However, the administration of all-trans retinoic acid carries known mutagenic toxic effects that are of concern when considering a vaccine for healthy children and women of childbearing age. Another potential limitation is that all-trans retinoic acid targets activated lymphocytes to the small intestine, but not necessarily to other mucosal surfaces such as the colon. Still, the study by Hammerschmidt et al. outlines in principle how the problem of mucosal immunization might be addressed. The approach warrants further investigation, but it would be premature at this point to abandon efforts to develop techniques for oral delivery of antigen and adjuvant for mucosal vaccination.

Disclosure forms provided by the authors are available with the full text of this article at NEJM.org.

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