

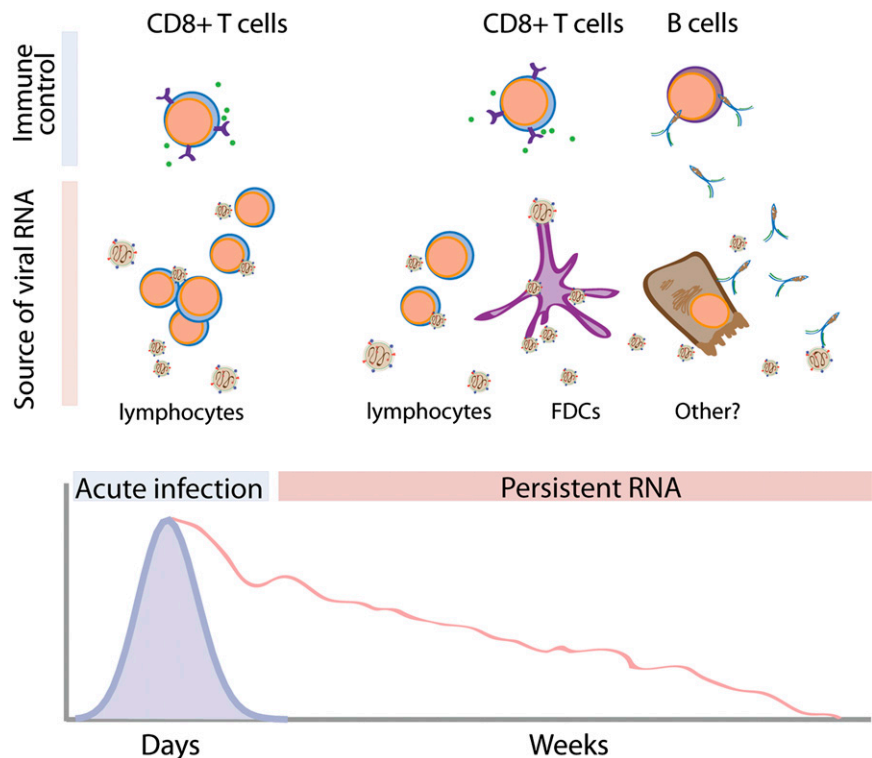
# Measles immunometrics

Theodore C. Pierson and Jonathan W. Yewdell<sup>1</sup>

Laboratory of Viral Diseases, National Institutes of Health, Bethesda, MD 20892

Like baseball, immunity is a team effort. Various innate/adaptive humoral/cellular components work in unison to clear infections. Determining the importance of each baseball player/immune element to winning/clearing is more difficult than meets the eye. Just as mathematical modeling of individual contributions (“sabermetrics”) revolutionized baseball, “immunometrics” will change our concept of immunity. Measles virus (MV) is a highly transmissible negative-stranded RNA virus that remains a major cause of childhood morbidity and mortality (1). Mortality results from MV infection itself or secondary bacterial infections facilitated by MV-mediated immunosuppression. Following the introduction of effective vaccines during the 1960s, worldwide annual MV mortality decreased from more than 2.8 million to less than 150,000. Vaccine efficacy, in conjunction with the apparent absence of an animal reservoir, potentially makes MV the second human pathogen (after smallpox) to be eradicated by medical intervention (2). MV infection elicits neutralizing antibodies (Abs) that correlate with lifelong measles protection (3). However, the clearance of MV from infected individuals was thought to be caused principally by CD8<sup>+</sup> T-cell activity. Infectious MV is detected in the blood by 7 d after exposure and is cleared within 2 wk, coincident with the appearance of T-cell responses (Fig. 1) (4–6). In both humans and monkeys, MV RNA can persist in the blood for months (7, 8), which may be related to MV-mediated immunosuppression, and, ironically, immunological memory as well. In PNAS, modeling MV immunity enables Lin et al. to propose a surprising role for Abs in mediating MV clearance (9).

Lin et al. infect rhesus macaques with MV via the respiratory tract, the normal route of viral transmission, and measure infectious MV and virion RNA (i.e., MV RNA) in blood. Infectious virus could be recovered only during the first ~14 d of infection. In contrast, MV RNA persisted for ~50 d, as described previously (7, 8). How does this correspond with the immune response? Three immune parameters are included in mathematical models predicting MV RNA dynamics. Neutralizing Ab titers are used as proxy for the humoral response. T-cell responses are inputted as the frequency of IFN- $\gamma$ -secreting cells in peripheral blood mononuclear cells. Finally, the immunosuppressive influence of



**Fig. 1.** The dynamics of MV clearance. MV infection results in transient viremia (purple line, *Lower*) that is cleared within ~2 wk, coincident with CD8<sup>+</sup> T-cell responses. MV RNA (red line, *Lower*) persists, however, for as long as months in blood. In the acute phase of infection, most MV is likely produced by infected lymphocytes. The source of persistent MV RNA is unknown, but could come from infected lymphocytes, follicular dendritic cells, or possibly other cell types, including epithelial cells in lung, liver, or kidney. Lin et al.'s modeling studies (9) point to an important role for Abs in clearing persistently infected cells that are the source of MV RNA.

regulatory T cells ( $T_{\text{regs}}$ ) is included as FoxP3 mRNA measured in peripheral blood mononuclear cells (FoxP3 is a transcription factor expressed at high levels by  $T_{\text{regs}}$ ). The total lymphocyte count is used as a measure of the number of MV susceptible cells in the animal.

By using relatively simple linear differential equations, the authors show that a model based solely on the contribution of effector T cells accounts for clearance of infectious virus, but not the persistence of MV RNA. Instead, and somewhat unexpectedly, accurately modeling MV RNA dynamics requires a major contribution from Abs (and, to a lesser extent,  $T_{\text{regs}}$ ). These findings elegantly illustrate the sequential coordination of cellular and humoral responses to fully clear MV from infected individuals.

CD8<sup>+</sup> T-cell-mediated clearance of infectious virus is consistent with these cells direct killing of infected cells and delivery of cytokines to induce innate antiviral im-

munity. However, how do Abs clear MV RNA? Answering this question requires knowledge of the nature and source of MV RNA. RNA degrades rapidly when unprotected by virions or other membrane-bound structures. MV RNA may therefore be released by persistently infected cells in the form of exosomes or virions that are not detected as infectious units as a result of their interaction with neutralizing Abs or insensitive culture methods. In this scenario, neutralizing Abs could contribute to clearance of persistent MV RNA by limiting cell-to-cell transmission of low levels of residual infectious virus. At the same time, Abs could play a direct role in eradicating infected cells

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<sup>1</sup>To whom correspondence should be addressed. E-mail: jyewdell@niaid.nih.gov.

via Ab-dependent cellular cytotoxicity or complement fixation (10). Less well defined, but still potentially relevant, is that Abs might induce antiviral activity in infected cells by direct interaction with viral proteins on infected cell surfaces (11).

The study of Lin et al. (9) highlights the importance of identifying the source of persistent MV RNA in infected individuals. Studies in primates with fluorescent protein-expressing MV demonstrate that the initial targets of MV infection are macrophages and dendritic cells of the upper respiratory tract (12). MV is amplified in regional lymph nodes and then disseminated hematogenously to multiple organs including lung, liver, and skin (where the telltale rash marks the T-cell response). Infected respiratory epithelial cells shed virus into the airway to enable transmission (13). Any or all of these cells could be the source of MV RNA, as MV RNA persists not just in blood, but also urine, lymphoid tissue, and lung secretions. MV can persistently infect neurons, spreading between cells directly without

budding (4). If MV is capable of similar tricks outside the CNS, this would certainly complicate Ab-mediated clearance.

On the contrary, if the prolonged persistence of MV RNA is accompanied by

## Modeling MV immunity enables Lin et al. to propose a surprising role for Abs in mediating MV clearance.

viral gene expression, this could contribute to the magnitude of the immune responses and, in particular, the remarkable duration of immunity. Is MV special in this respect? Recent findings in mice infected with “acute” viruses demonstrate a similar surprising duration of viral RNA, which can last for years (14) and be accompanied by viral immunogens capable of activating

CD8<sup>+</sup> T cells (15, 16). It would certainly make sense for the immune system to retain the genetic information of previous viral infections to maintain memory by occasionally translating retained viral mRNA, although it is clear that antigen persistence is not an absolute prerequisite for maintaining memory T cells (17).

Immunometrics is a powerful approach for deconvoluting the contribution of distinct components of the immune response to infection. As exemplified by Lin et al. (9), immunometrics tests assumptions and raises important questions for further study. At the same time, much remains to be learned on the road to accurate modeling. Above all, we should be ever-vigilant of the danger of immune correlates masking true effector functions. A model that matches the data is not equivalent to a model that explains the data. This is an intellectual trap. Indeed, as Lin et al. show (9), the safer scenario is when models fail, as this paves the path to new testable hypotheses.

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