

**5 Estimating Vaccine Efficacy with Unequal Follow-up of Vaccinated and Unvaccinated Groups**

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**Objectives:** Develop a vaccine efficacy estimator that can accommodate various types of unequal follow-up in vaccinated and unvaccinated groups.

**Background:** Vaccine efficacy (%) has been estimated by comparing attack risks in vaccinated (VR) and unvaccinated (UR) groups in the following manner:  $VE=100*(1-(VR/UR))$ . The underlying biological model is that the vaccine produces complete immunity in a percentage of those vaccinated (VE) while the rest remain as susceptible to disease as those unvaccinated. This estimator is limited, however, to the situation where there is equal (potential) follow-up of all study participants. That is, the estimator does not allow: 1) loss to follow-up (i.e., censoring), 2) late entry to study, or 3) switching from unvaccinated to vaccinated (e.g., vaccination in a disease outbreak).

**Methods:** We calculate the probability that a vaccinated individual remains susceptible after vaccination based on the observed follow-up period and the pattern of cases that occur during the follow-up period. These probabilities are combined to provide a vaccine efficacy estimator. Taylor series expansions are used to obtain a valid variance for the estimator. The properties of the new estimator are evaluated in simulations and existing measles epidemic data sets.

**Results/Conclusions:** In situations with equal follow-up the two methods provide nearly identical results. Simulation results indicate that the new estimator does provide an unbiased and appropriate basis for estimating vaccine efficacy with unequal follow-up of vaccinated and unvaccinated groups.

**7 DNA Immunization Protects Nonhuman Primates Against Rabies Virus**

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Worldwide, approximately 50,000 people die annually from rabies with most fatalities occurring in developing countries where rabies is endemic and health-care delivery is inadequate. Many countries still use vaccines produced in sheep, goat or suckling mouse brain because cell culture-based vaccines are expensive. The stability and low cost for mass production of DNA vaccines would make them ideal for use in developing countries. To determine the potential of DNA vaccines as a human rabies biologic, *Macaca Fascicularis* (*Cynomolgus*) monkeys were vaccinated with DNA encoding the glycoprotein of the challenge virus standard rabies virus, or with a human diploid cell vaccine (HDCV). The monkeys then were challenged with a non-passaged street rabies virus. DNA or HDCV vaccination elicited comparable primary and anamnestic neutralizing antibody responses. All 10 vaccinated monkeys (DNA or HDCV) survived rabies virus challenge, whereas monkeys vaccinated with the vector alone, developed rabies. Furthermore, sera from DNA or HDCV vaccinated monkeys neutralized a global spectrum of rabies virus variants. This study showed that DNA immunization elicits protective immunity in nonhuman primates against lethal challenge with a human viral pathogen of the central nervous system.

**6 Recruitment for Large Vaccine Trials at Individual Investigative Sites**

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**Objective:** To systematize the process of and predict the required resources and outcome for rapid participant accrual at an individual site for a large scale vaccine trial.

**Methods:** A recruitment plan for two related influenza vaccine studies was developed and funded by the study sponsor. Target enrollment was 600 in 10 weeks. A mailer was sent to 30,000 patients of a large medical group. Radio and print ads, local flyers and neighborhood canvassing supplemented the mailing. A call center screened and tracked calls, appointments and enrollees by recruitment method and cost. These data were used in real time to allocate resources to each recruitment method.

**Results:** Numbers of Calls and Enrollees and Cost per Unit by Recruitment Method

	Mailers		Radio		Flyers		Canvas/Other		Totals	
	No.	\$/Unit	No.	\$/Unit	No.	\$/Unit	No.	\$/Unit	No.	\$/Unit
<b>Calls</b>	585	\$27	461	\$28	40	\$3	209	N/A	1351	\$21.21
<b>Enroll</b>	290	\$54	239	\$54	49	\$2	73	N/A	651	\$44.02

Over 10 weeks, 1351 calls resulted in 651 enrollees. The mailing and radio ads were equally effective. Ratios of mailer number and radio ads to call and enrollee number became apparent after the first 200 enrollees. These ratios predicted the time and cost required to complete enrollment using these methods.

**Conclusions:** The time and cost for large-scale vaccine trial enrollment can be predictably managed at the site level. Success requires a plan with multiple recruitment methods, tracking results in real time, and selecting the most effective and least expensive methods accordingly. Identification of large populations of accessible eligibles is critical. We believe that each plan must be individualized and that *all successful recruitment is local*.

**8 Protective CTL Responses Against Paramyxoviruses Induced by Epitope-Based DNA Vaccines**

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**Objective:** To develop epitope-based DNA vaccines and to test their ability to induce protective immune responses against infection with measles virus (MV) and respiratory syncytial virus (RSV).

**Methods:** Plasmid DNA vectors were constructed with minigenes encoding a single cytotoxic-T-cell (CTL) epitope from either the M2 protein of respiratory syncytial virus (RSV) or from the nucleoprotein of measles virus (MV) with or without an immunoglobulin kappa chain signal sequence. <sup>51</sup>Cr release assay was used to measure CTL activity. Protection studies in murine models for MV and RSV were performed in CBA and BALB/c mice. *In vivo* depletion of IFN- $\gamma$  in mice was administered using monoclonal anti-IFN- $\gamma$  antibodies.

**Results and Conclusions:** Following intradermal immunization, plasmids in which the CTL epitopes were expressed in frame with the signal sequence, were more effective at inducing peptide- and virus-specific CTL responses than plasmids expressing CTL epitopes without the signal sequence. This immunization resulted in protection against MV-induced encephalitis and a significant reduction in viral load following RSV challenge. The reduction of viral load following RSV challenge was abrogated by prior injection with anti-IFN- $\gamma$  antibodies. These results highlight the potential of this approach for the development of vaccines against infectious diseases.