Practice of Epidemiology

Estimating the Severity Profile of Enterovirus A71 Infections in Children: A Bayesian Synthesis Framework

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Enterovirus A71 (EV-A71) is responsible for the majority of severe cases of hand, foot, and mouth disease, but little evidence is available on the severity profile of EV-A71 infections. We formulated a hierarchical Bayesian model that synthesized data on diseases/events associated with EV-A71 and EV-A71 antibody responses to infection among unvaccinated children from large clinical trials of EV-A71 vaccination, which were conducted in Jiangsu and Beijing during 2012 and 2013, to reconstruct the severity profile in a unified framework. On average, 15.1% of the children aged 6–35 months were infected by EV-A71 during 1-year follow-up in a mild epidemic season. We estimated that 9.7%, 2.2%, and 0.6% of children infected with EV-A71 were diagnosed with EV-A71-associated diseases, were hospitalized, and showed severe complications, respectively. We estimated on average 1 death per 10,000 EV-A71 infections for children aged 6–35 months. Approximately 70% of children had ≥ 4 -fold rises in antibody titers after infection. Most EV-A71 infections in young children are mild, and overall 2.2% of the infected patients were hospitalized in the 2 trials. There remain several uncertainties about the immune response after infection and the duration of immunity against EV-A71 reinfection.

Bayesian inference; EV-A71 infection; infection-fatality risk; severity profile

Abbreviations: CrI, credible interval; EV-A71, enterovirus A71; GMT, geometric mean titer; HFMD, hand, foot and mouth disease.

The clinical severity pyramid reflects the concept that for many infections, only a small fraction causes severe disease. A larger fraction might not be serious enough to require clinical attention, and a substantial fraction might be subclinical. The clinical severity pyramid has been characterized for some infectious diseases (1, 2), and one commonly used measure of the severity profile of an acute infectious disease is the case-fatality risk. Because of variation in health-care seeking behaviors and in the selection of cases for testing, the infection-fatality risk could be a more stable measure of severity than the case-fatality risk (3). Several studies have attempted to infer the virus infections from various types of serological data, such as community-based studies (4) and cross-sectional seroprevalence studies that collected serum from blood donors (5) or diagnosed patients (6), while evidence from serum randomly sampled from the general population is not available at present.

Enterovirus A71 (EV-A71) is of particular interest as a major causative pathogen of severe and fatal hand, foot, and mouth disease (HFMD), which is a self-limiting infectious disease that

is common in young children in Asia (7–9). Serological studies have shown that the maternal immunity against EV-A71 infection decays at approximately 5 months of age, and the majority of children have had EV-A71 infection by 4 years of age (8–10). A small proportion of HFMD patients suffer from severe neurological complications, such as meningitis and encephalitis, and the majority of these severe complications are associated with EV-A71 infections (7). No severity profile has yet been characterized for EV-A71 infections, while the estimates of the case-fatality risk of EV-A71 cases vary from 0.4% to 7.9% (11, 12). Two candidates for monovalent EV-A71 vaccines have been assessed in phase-3 trials and have been licensed in China, providing 2 sets of serum profiles randomly collected from the high-risk population of EV-A71 infection (i.e., children aged 6–35 months).

The objective of this study was to characterize the severity profile of EV-A71 infections in children. We applied a hierarchical Bayesian model, which synthesized published evidence from vaccine trials and from epidemiologic and serological

studies to estimate various quantities of severity. In particular, the use of published serological correlates of protection allowed the estimation of asymptomatic and symptomatic infections where a population-based serological study was unavailable. The hierarchical model was initially applied to the data set from each trial separately. An extended joint model was then applied to both data sets simultaneously, and we report these results as the main results. Although the pooled estimates with improved power can be estimated from the joint model, we nevertheless expected that the force of infection, initial population susceptibility, and proportion of severe events would be heterogeneous due to the different background HFMD epidemiology as well as different intensity in the follow-up of patients across trial sites.

METHODS

Sources of data

Data on EV-A71-associated events and the prevalence of different titers of antibodies against EV-A71 among healthy children aged 6–35 months were obtained from the published results for unvaccinated children (placebo arm) reported by 2 phase-3 clinical trials (Sinovac and Vigoo) of EV-A71 vaccine candidates, which were conducted in 2012–2013 in Beijing and in Jiangsu municipality/province in China (13, 14). Samples of 5,028 and 5,115 children aged 6–35 months were enrolled in the placebo arms of the Sinovac and Vigoo trials, respectively (13, 14). EV-A71-associated events (i.e., medically attended symptomatic disease, hospitalization, severe complications, or death) were defined as the simultaneous presence of the above clinical events and evidence of EV-A71 infection, which was defined by either the successful isolation of EV-A71 virus or no less than 2 consecutive positive results for EV-A71 RNA (13, 14). Symptomatic disease includes HFMD, herpangina,

and/or other illnesses, and severe disease includes neurological or other severe complications (13, 14).

The titer distribution and geometric mean titer (GMT) for both unvaccinated children (referred to here as “all children”) and unvaccinated children who were seronegative to EV-A71 antibody (titer <8) at baseline only (referred to as “baseline-seronegative children”) were obtained from the trial reports. Neutralizing antibodies against EV-A71 were measured by the modified cytopathogenic effect assay, with the dilution starting at 1:8 and increasing by a factor of 2 (13, 14). Information on EV-A71 antibody titers was available for 571 (Sinovac) and 620 (Vigoo) children who enrolled in the immune cohort from the placebo arms (13, 14). Antibody titers less than 8 were imputed as 4 for calculation purposes. The 1-year infection risk was inferred from the antibody titers measured at the start (56 days after baseline) and end (14 months after baseline) of the follow-up periods. Web Table 1 (available at <https://academic.oup.com/aje>) summarizes the data used in this work. In addition, we extracted information on the case-fatality risks of HFMD cases caused by EV-A71 in China from the report by Xing et al. in 2014 (15) and the serological correlates of protection against EV-A71-associated disease from the report by Jin et al. in 2016 (16).

Statistical model

A Bayesian model was formulated to estimate the incidence of infections and severity risks of EV-A71 infection among children (Figure 1) by adapting previous work (1, 17). We assumed 4 levels of severity after EV-A71 infection (I), which in the order of severity were: medically attended symptomatic cases (S), hospitalization (H), severe complications (V), and death (D). Children progressing to a more severe disease level m were modeled as a subset from the previous level l , following a binomial distribution with probability p_{ml} (Figure 1 and Table 1).

Figure 1. Statistical framework to estimate the severity risks of enterovirus A71 (EV-A71) infection. A) Progression of EV-A71 infection among unvaccinated children. Double circles represent the number of events that were not directly available. Solid arrows indicate the conceptual pathway of the framework, and dashed arrows indicate the parameters estimated in the model. The probability of children infected by EV-A71 ($p_{I|N}$) was inferred from the parameters in the second panel (B). A titer-dependent infection model was used to estimate the incidence of EV-A71 infections.

Because EV-A71 infections can be mild, serological data were used to estimate the number of children infected with EV-A71 irrespective of clinical presentation (Figure 1B). We estimated the number of EV-A71 infections based on the observed distribution ($n_{T_i,t,k}$) of different titer levels ($T_{i,t,k}$) indexed by i ($T_i = 2^{i+1}$, $i = 1, 2, \dots, 10$) and GMT ($G_{t,k}$) at time point t (beginning or end) of the follow-up for group k (all children or baseline-seronegative children), after accounting for titer-dependent protective effects against EV-A71 infection (p_{IT_i}) and immunity boosting ($p_{T_i \rightarrow I}$). In brief, $N_{t,k}$ children were observed from group k ($k = 1$ for “all children” and $k = 2$ for “baseline-seronegative children”) in the placebo arm at time t . We assumed the GMT followed a log-normal distribution. The observed number ($n_{T_i,t,k}$) of children with titer T_i was modeled to follow a binomial distribution with probability $p_{T_i,t,k|N_{t,k}}$ and denominator $N_{t,k}$. We assumed that the number of children who had antibody titer of ≥ 32 ($i = 4, 5, \dots, 10$) decreased exponentially with titer at a rate ρ , given that the specific titer values for these children were unavailable.

A previous study, based on a susceptible-infectious-recovery transmission model, suggested that stratified immunity provides better predictions of the infections than does a binary seropositivity assumption (17). The probability of children infected with EV-A71 in all titer groups was modeled as follows:

$$p_{IN_{1,k}} = \sum_i p_{IT_i} p_{T_i,1,k|N_{1,k}}$$

A higher EV-A71 antibody titer was assumed to be associated with a lower risk of EV-A71 infection, which was modeled by the probability (p_{IT_i}) of infection given titer T_i . A previous study examined the serological correlates of protection against disease associated with EV-A71 by estimating the probability of disease exposure (parameter λ) and the logit scaled protection of titer (depending on parameters α representing the location and β representing the scale of protection) (Table 1) (16). We modeled the association as follows:

$$p_{IT_i} = \frac{\lambda}{(1 + e^{\alpha + \beta \log(T_i)}) p_{SI}}$$

where p_{SI} denotes the infection-symptomatic risk.

We assumed that the boosting in antibody titers after infection followed a Poisson distribution with an average 2^μ -fold rise, where the parameter μ was estimated. Taking account of the probability of EV-A71 infection from a child's own titer group and the immunity boosting from lower titer groups, the probability of a child being in titer group i at the end of follow-up was:

$$p_{T_{i,2,k}|N_{2,k}} = (1 - p_{IT_i}) p_{T_{i,1,k}|N_{1,k}} + \sum_j p_{T_{i-j} \rightarrow T_i|I} p_{IT_{i-j}} p_{T_{i-j,1,k}|N_{1,k}},$$

where $p_{T_{i-j} \rightarrow T_i|I}$ represents the probability of a child in titer group i moving to group $i + j$ after infection ($j = 1, \dots, 10$).

After applying the above hierarchical model to data sets from each trial separately, we extended our model to derive a set of joint estimates of probability of severity risks following EV-A71 infections, while allowing for heterogeneity of initial susceptibility of the local population and disease prevalence (see Web Appendix 1). In our main analysis, we assumed that immunity against EV-A71 would not decay during a 1-year period, and we performed sensitivity analyses in which antibody titers decayed to one-half or one-fourth of their original levels, respectively, across a 1-year period. We conducted sensitivity analyses of different assumptions of the titer distribution in the group with titer of ≥ 32 . We also conducted a sensitivity analysis in which the proportion of EV-A71 infections was derived by defining EV-A71 infections as children with antibody titers of ≥ 8 , 16, or 32, to examine the potential uncertainty of our estimates. We used data on the case-fatality risk of EV-A71 in mainland China to inform the prior distribution of p_{DIS} (15). Due to a lack of individual data on EV-A71 antibody titers, the estimates of the protection associated with antibody titers against EV-A71 infection among unvaccinated children were

Table 1. Parameters for a Model to Estimate Incidence of Infections and Severity Risks Among Cases

Parameter	Description	Prior Distribution	Reference
$p_{T_i,1 N_1}$	Probability of children in antibody titer group i at start of follow-up ($i = 1, 2$, or 3).	Uniform (0, 1)	
ρ	Parameter to determine the titer distribution of the ≥ 32 -titer group.	Uniform (0, 10)	
α	Location parameter of the probability that a child was protected by titer t .	Normal (−1.84, 3.78)	Assumption (13)
β	Scale parameter of the probability that a child was protected by titer t .	0.71	Assumption (13)
λ^a	Probability that a susceptible individual suffered from EV-A71-associated disease.	Sinovac: 0.041 Vigoo: 0.017	Assumption (13)
μ	Mean 2^μ -fold of boosting after EV-A71 infection.	Uniform (0, 20)	
p_{SI}	Probability of a child showing symptoms after EV-A71 infection.	Uniform (0, 1)	
p_{HIS}	Probability of a medically attended symptomatic case being hospitalized.	Uniform (0, 1)	
p_{VIS}	Probability of a medically attended symptomatic case having severe complications.	Uniform (0, 1)	
p_{DIS}	Probability of death of a medically attended symptomatic case.	Normal (0.001, 9×10^{-8})	Assumption (12)

Abbreviation: EV-A71, enterovirus A71.

^a Data was obtained from the published results for unvaccinated children (placebo arm) reported by 2 phase-3 clinical trials (Sinovac and Vigoo) of EV-A71 vaccine candidates (13, 14). For Sinovac, λ was scaled by the incidence ratio between Sinovac and Vigoo, $\lambda_{Sinovac} = \lambda_{Vigoo} \frac{I_{Sinovac}}{I_{Vigoo}}$.

used to inform the prior distribution of α (15, 16), while estimated values for β and λ among unvaccinated children were extracted from a previous study, which was based on the Vigoo trial (16). We assumed different values for β and λ as a sensitivity analysis to examine their potential impacts on our estimation. The probability of a susceptible individual suffering from EV71-associated disease (λ) was highly affected by local disease prevalence and therefore was varied across locations. We scaled the value of λ from the Sinovac trial by the incidence ratio between the 2 trials (Table 1 and Web Table 2). Uniform prior distributions were assumed for all other parameters (Table 1). The model was fitted to the data from the 2 trials separately. We used a Monte Carlo Markov chain method using the Metropolis-Hastings algorithm implemented in R, version 3.4.2 (R Foundation for Statistical Computing, Vienna, Austria), to estimate the parameters, where 100,000 samples were drawn from the posterior distribution of each parameter after removing 50,000 burn-in iterations. The deviance information criterion was calculated to aid model selection (18). We present the estimated infection risks of EV-A71 and other outcomes per 1,000 infections.

Simulations

To test the validity of the statistical framework, we first simulated 2 sets of stochastic realizations of individual data, with 100 realizations for each set. Scenario 1 was simulated by assuming conditions similar to the Vigoo trial, while scenario 2 assumed a higher disease prevalence and a more susceptible population (Web Table 4). We then applied the hierarchical model to the aggregated data from each realization and compared the model estimates with the simulated observations. We also compared the observed EV-A71-associated severe outcomes and antibody titers versus 5,000 predictions drawn from the posterior predictive distribution from our main analysis for both trials. In order to examine the impact of overdispersion in the antibody titer-boosting distribution, we carried out a sensitivity analysis under scenario 1 but assumed a negative binomial distribution (mean = 2.4, variance = 3.6 (1.5 times that in the main analysis)) for the immunity boosting. All analyses were conducted in R, version 3.4.2 (R Foundation for Statistical Computing).

RESULTS

Results from analyses of the 2 trial data sets indicated that, overall, 15.1% (95% credible interval (CrI): 13.7, 16.5) of children 6–35 months of age were infected by EV-A71 during 1 year of follow-up (Table 2). Overall, 9.7% (95% CrI: 8.1, 11.4) of EV-A71-infected children showed symptoms and were diagnosed with EV-A71-associated HFMD or other related diseases. On average, 2.2% of EV-A71-infected children were hospitalized in the 2 trials, while fewer than 1% of children developed severe complications after EV-A71 infections. We estimated that there was on average 1.0 (95% CrI: 0.6, 1.4) death per 10,000 EV-A71 infections among children aged 6–35 months (Table 2).

Results from the simulation analysis with similar conditions to the Vigoo trial (scenario 1) suggested that 87% of the

credible intervals covered the true values of the proportion of EV-A71 infections, while all the true values of the proportions that were medically attended and hospitalized after EV-A71 infections were covered (Figure 2). The credible intervals of the proportion of children who developed severe complications after EV-A71 infections covered all true values when they were nonzero. Overall, the estimates captured the variation in various estimates of the severity profile (Figure 2). The models also, in general, had good predictions compared with simulated data under conditions different from the Vigoo trial (see Web Figure 1) and the observed data from the 2 phase-3 trials (see Web Figure 2).

Results from the sensitivity analysis suggested a limited effect of the assumed titer distribution in the ≥ 32 -titer group on model performance, while the models assuming no decay of EV-A71 immunity generally showed better performance than models assuming decay in EV-A71 immunity (see Web Table 3). The estimated proportions of EV-A71 infections among the studied populations increased slightly with the assumption of a higher rate of decay in immunity (Figure 3). The estimated risks of EV-A71-associated events were similar across models (Figure 3). Slightly higher severity risks after infection and lower proportion of EV-A71 infections among the studied children were estimated by models assuming higher values for both β and λ , but these estimates were still very similar to those from the main analysis (see Web Figure 3). Smaller proportions of EV-A71 infections and larger proportions of medically attended cases were estimated from the models defining EV-A71 infections by a higher cutoff of antibody titer (see Web Figure 4).

Results from the main analysis estimated that antibody titers rose by factors of 6.2 (95% CrI: 5.0, 7.9) for the Sinovac trial and 5.4 (95% CrI: 4.0, 7.2) for the Vigoo trial after EV-A71 infection, while 74.0% (95% CrI: 67.5, 79.9) and 70.1% (95% CrI: 61.3, 77.7) of the children, respectively, were estimated to experience at least a 4-fold increase in antibody titers against EV-A71. Higher proportions of infected children having a ≥ 4 -fold rise in EV-A71 antibody titers were estimated from the models that allowed for decay in EV-A71 antibody titers compared with the models assuming no decay in antibody titers (Figure 4). Results from the sensitivity analysis suggested that overdispersion in antibody boosting seems not affect our estimation on the severity profile (Web Figure 5) and estimated level of boosting (6.2, 95% CrI: 4.4, 8.9).

DISCUSSION

During 1 year of follow-up in the placebo arms of 2 EV-A71 vaccine trials, an estimated 15.1% of the children aged 6–35 months were infected with EV-A71. Both study locations experienced a relatively low level of EV-A71 activity in 2012–2013 (19, 20), so the estimates of infected proportion might be lower than those in a normal epidemic year. We also applied age-specific seroprevalence of EV-A71 antibodies estimated by a previous meta-analysis to the corresponding age groups in China (10), and we estimated a similar proportion (8.4%) of EV-A71 infections among children aged 6–35 months across the country during 2008 and 2013. A previous study in Cambodia suggested that the annual risk of EV-A71 infection

Table 2. Estimated Number and Risk of Events Associated With Enterovirus A71 Infections, Beijing and Jiangsu, China, 2012–2013^a

Associated Event	Sinovac ^a		Vigoo ^a		Overall ^b	
	Posterior Median	95% CrI	Posterior Median	95% CrI	Posterior Median	95% CrI
Infections, per 1,000 children	178.7	159.3, 198.9	115.9	99.8, 133.1	150.5	137.3, 164.5
Medically attended cases, per 1,000 infections	118.7	95.3, 145.9	69.8	49.8, 94.9	96.7	81.1, 113.5
Hospitalizations, per 1,000 infections	27.5	17.9, 40.4	14.6	6.9, 27.1	21.5	14.8, 30.1
Severe complications, per 1,000 infections	9.6	4.5, 18.0	2.8	0.4, 9.4	6.4	3.1, 11.4
Deaths, per 10,000 infections	1.2	0.7, 1.8	0.7	0.4, 1.1	1.0	0.6, 1.4

Abbreviation: CrI, credible interval.

^a Estimated from the hierarchical model applied to the data set of each trial separately. Data was obtained from the published results for unvaccinated children (placebo arm) reported by 2 phase-3 clinical trials (Sinovac and Vigoo) of enterovirus A71 vaccine candidates (13, 14).

^b Estimated from the hierarchical model applied to both data sets of the trials simultaneously.

fluctuated in the range of 5%–75% during 1994–2011 for children aged 2–15 years (6). The estimates of our study were similar to their estimates from mild epidemic seasons.

Our results suggested that, on average, 9.7% of the EV-A71 infections were symptomatic, required medical attention, and were diagnosed with HFMD or other related diseases. A recent

Figure 2. Observed and estimated proportion of enterovirus A71 infections and risk profile for developing more severe forms of the disease among infected cases from the simulation analysis. The data was simulated assuming conditions similar to those in the Vigoo trial (13). The number of infections is per 1,000 children, and the number of events is per 1,000 infections.

Figure 3. Comparison of estimates of enterovirus A71 infections and infection risks from models using different assumptions about the rate of decay in immunity, using data from the Sinovac and Vigoo trials, China, 2012–2013 (13, 14). The number of infections is per 1,000 children, and the number of events is per 1,000 infections. Grey represents estimates from the main analysis. For data from children with antibody titers of ≥ 32 , blue dashed lines represent estimates from models assuming the numbers were evenly distributed, and blue dotted lines represent estimates from models assuming that they were exponentially increasing. Red dashed lines show estimates from models assuming antibody titers decayed to one-half of the original level, and red dotted lines show estimates from models assuming antibody titers decayed to one-fourth of the original level.

Figure 4. Estimates of the proportion of children with 4-fold or greater rises in antibody titers after enterovirus A71 infection using different assumptions about the rate of decay in immunity, using data from the Sinovac and Vigoo trials, China, 2012–2013 (13, 14). Grey represents estimates from the main analysis. For data from children with antibody titers of ≥ 32 , blue dashed lines represent estimates from models assuming the numbers were evenly distributed, and blue dotted lines represent estimates from models assuming that they were exponentially increasing. Red dashed lines show estimates from models assuming antibody titers decayed to one-half of the original level, and red dotted lines show estimates from models assuming antibody titers decayed to one-fourth of the original level.

study in Beijing found that medical attendance for children infected with EV-A71 was driven mainly by the appearance of rash; however, only 10.7% of EV-A71 seropositive children developed rash (21). An earlier study in Taiwan estimated that 29% of EV-A71-seropositive preschool children were symptomatic (i.e., with herpangina or HFMD and developing rash) (22). With only a portion of these children developing rash, the proportion of infected children seeking medical attention should be similar to our estimate. In our previous studies, we also estimated that an average of 15.8% of EV-A71-infected children aged 6–35 months in China were reported to the national HFMD surveillance system, using the EV-A71 seroprevalence and EV-A71-associated HFMD reported previously (7, 10). In a Shanghai study, among children who had EV-A71 antibody titers greater than 8, 9.8% of the parents recalled that their children had a previous diagnosis of HFMD (23). This proportion is comparable to the estimated 9.7% symptomatic proportion in our study, although in the Shanghai study any HFMD diagnoses over the previous years were considered (age range, 0–5 years). Two studies from Taiwan reported high proportions of symptomatic EV-A71 infections, 71.4% and 67.6%, respectively (24, 25). This is probably because the criterion of a 4-fold or greater rise of EV-A71 neutralizing antibody in paired sera might have led to an underestimated number of infections, given that our results suggested that approximately 20%–30% of children infected with EV-A71 have less than a 4-fold rise in antibody titers, and these children are probably less likely to have symptoms (Figure 4).

We estimated that an average of 2.2% of EV-A71-infected children aged 6–35 months were hospitalized, and 0.6% of

EV-A71-infected children suffered from severe complications, suggesting that only 29% of EV-A71 hospitalizations were associated with severe complications. This was fairly consistent with a previous study in which the majority of hospitalized HFMD patients did not suffer severe complications (26). Our estimated proportions of medically attended and hospitalized cases following EV-A71 infections seem to suggest noticeable heterogeneity across the 2 trials (Table 2, Web Figure 6, and Web Tables 5 and 6). One possible reason is that these proportions depend not only on disease progression but also on factors such as health-seeking behavior, health insurance status, or hospital admission criteria (27). Hence, the pooled estimates of the severity risks should be interpreted with caution given the potential heterogeneity across settings.

Our results revealed several uncertainties about the immunity against EV-A71 virus. First, the level of immunity boosting after EV-A71 infection was poorly understood. We estimated that EV-A71 antibody titer rose by an average of about 5–6 times after infection. A 4-fold or greater rise in EV-A71 neutralizing antibody titer in the paired sera has been widely applied to detect past infection of EV-A71 (9, 13). However, our estimated proportion of children whose EV-A71 antibody titer increased 4-fold or more after infection ranges from 70% to 80% across models, suggesting that a fraction of EV-A71-infected children might not be identified under the usual criteria. This is also supported by previous serological findings suggesting that approximately 19% of diagnosed HFMD cases did not have an EV-A71 antibody titer ≥ 16 after infection, and 6.8% of HFMD patients were seronegative (28). In addition, our predictions on GMT were slightly lower than observed. This was probably because our model assumed the highest value for antibody titer was 2,048, but the observations might include serum from patients in the acute recovery phase. Immunoglobulin M titer is usually much higher than immunoglobulin G titer in the acute recovery phase, but neutralizing antibody tests were not able to differentiate between immunoglobulin M and immunoglobulin G (28); therefore the extremely high immunoglobulin M titer could have inflated the observed GMT of EV-A71 antibodies.

Second, the long-term persistence and protective effects of immunity against EV-A71 still require further research. In the sensitivity analysis, we compared models with different assumptions on the decay pattern in EV-A71 antibody titers and found a better performance for the models assuming no decay in titers (see Web Table 2). This might suggest a limited decay of antibody titers against EV-A71 within 1 year of follow-up. To our knowledge, no prospective study has been conducted to examine the long-term dynamics of EV-A71 immunity. The protective effect of neutralizing antibody against EV-A71 was assumed to increase with higher titer level, following a scaled logit model (16), while the values of β and λ seem to have limited impact on our main conclusion (see Web Figure 3). These serological correlates were validated by a cohort of children aged 6–35 months, and the generalizability of the serological correlates was not clear (see Web Figures 3 and 6). Additionally, the antibody titer increase was discretized to factors of 2 in both of the original trials, and we were not able to quantify the uncertainty associated with such discretization using aggregated data.

Our study has several limitations. First, our estimates of the parameters related to EV-A71 immunity dynamics might be less accurate due to lack of specific data on the titer values of children with EV-A71 antibody titers of ≥ 32 . However, results from the sensitivity analysis suggested that the missing data on titer distribution appear to have limited impact on our final estimates, probably because about 90% protection against EV-A71 infection would be provided by an antibody titer of ≥ 32 , according to our estimates and previous studies (14, 16). Besides, we sampled with equal probability the children with antibody titers of ≥ 32 in the simulation analysis and applied a model assuming an exponential distribution for the decreasing proportions of individuals with higher titer levels. The results again suggested the limited impact of the missing data on our final estimates, even when a lower protection of titer against infection was assumed (Figure 2 and Web Figure 1). Second, we were not able to compare our estimates of EV-A71 infections with the gold standard of EV-A71 detection (at least 2 consecutive, successful virus isolations) because virus isolation was conducted only for diagnosed symptomatic cases. Third, the data we used in this study was collected mainly from Jiangsu and Beijing during a milder epidemic season in 2012–2013, and the estimated risk of infection might not be generalizable to other parts of mainland China or other countries. We estimated the cumulative incidence of EV-A71 infections and symptomatic infection risk using previously reported serological and notification data, suggesting a similar range with our main estimates (7, 10). Finally, we estimated an infection-fatality risk of 1.0 per 10,000 for EV-A71 infection among children aged 6–35 months. However, because no fatal cases were reported in the vaccine trials, the estimation of infection-fatality risk was informed largely by the risk of death among clinically diagnosed EV-A71-associated HFMD cases reported to the national surveillance system (15).

In this study, we applied a hierarchical Bayesian framework to synthesize evidence from phase-3 trials of EV-A71 vaccines and characterize the severity profile of EV-A71 infections among the general population at risk. Our model provided good estimates, especially when the observed number of events was nonzero. The unified framework might also take into account uncertainties from different data sources to provide estimates with appropriate associated uncertainties for various quantities of interest. Previous attempts to outline the complete range of clinical severity were often challenged by the uncertainty in the number of infections in the population (1). Our model used aggregated data on the serological correlates of protection against infection and the immune response and observations of the infection-associated diseases, which are routinely reported by vaccine clinical trials. Prior information on the case-fatality risk might also be needed when there are few observed deaths in the trials. Our model can be used to obtain a plausible profile for the risk of developing more severe forms of the disease among infected cases, which is critical to understanding the disease burden, transmission dynamics, and potential impact of vaccine in the population. Further adaptation of the model to differentiate the immune response between the first and subsequent infections could be helpful, especially for viruses that can cause repeated infections (29). Our approach is applicable to a wide range of diseases, especially when population-based serological study is not

available, to provide the complete severity profile from sub-clinical infections to death.

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