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Pre-vaccination evolution of antibodies among infants 0, 3 and 6 months of age: A longitudinal analysis of measles, enterovirus 71 and coxsackievirus 16

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Abstract

Background—Due to waning levels of maternal antibodies (measles; enterovirus 71, EV71; and coxsackievirus A16, CoxA16), some infants may lose protection against infection prior to vaccination. Using a longitudinal design, we examine how maternal antibody levels evolve over time in infants prior to vaccination.

Methods—In 2013–2014, we collected sera at ages 0, 3 and 6 months from infants. We assayed for levels of measles IgG antibody (717, 233 and 75 sample sera tested at months 0, 3 and 6, respectively), and neutralizing antibodies for EV71 and CoxA16 (225, 217, and 72). Demographic and health information were collected, and a linear mixed model (LMM) was used to describe antibody levels over time.

Results—Pre-vaccination monotonic antibody decreases were observed for measles (1410, 195 and 22 mIU/ml, $p < 0.001$), EV71 (1:19.9, 6.3 and 4.5, $p < 0.001$) and CoxA16 (1:16.3, 5.9, and 4.5, $p < 0.001$). At 6 months of age, only 2.7% (95%CI, 0.6–8.3), 6.8% (95%CI, 2.7–14.4) and 5.6% (95%CI, 1.9–12.7) of infants were antibody positive for measles, EV71 and CoxA16, respectively. LMM findings indicated that infants with higher antibody titers at birth experienced a greater loss of antibody level. An infection rate of 1.3% (95%CI, 0.1–6.1) was reported for both EV71 and CoxA16.

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Author contributions

C.F. and M.W. designed the study; L.L. collected the serum samples; Y.L. and Y.C. did the experiments; Q.G., S.P. and Z.Y. built the dataset; C.F. and J.S. analyzed the data and C.F., Q.G. and J.S. wrote the manuscript text.

Competing interests

JS discloses partial ownership of SK Analytics.

Conclusions—Further modifications of vaccination strategies for measles, earlier vaccination for EV71 infection, and deployment of a CoxA16 vaccine need to be considered to limit infection among the very young.

Keywords

Maternal antibodies; Infants; Measles; Enterovirus 71; Coxsackievirus 16

1. Introduction

Specific maternal antibodies provide vital, passive immunity against infectious pathogens for infants during the first months of life [1]. However, depending on an infant's immunization schedule, this maternal protection may wane prior to vaccination [2–4]. Over the past decade, measles and hand-foot-mouth disease (HFMD) have remained public health challenges among infants in some countries, including China [5,6]. This phenomenon may, in part, be due to the timing of immunization.

Measles vaccination is free and mandatory for children 8 months to 14 years of age, and since vaccination began in China, reported measles cases have decreased substantially. Indeed, mean annual measles incidence was 572.0 per 100,000 between 1960 and 1969, 355.3 per 100,000 between 1970 and 1979, 52.9 per 100,000 between 1980 and 1989, and 7.6 per 100,000 between 1990 and 1999 [7]. Since 1986, the control of measles incidence in China has been supported by the Expanded Program on Immunization (EPI), which provides measles vaccination using a two-dose successive vaccination schedule. In the 30 years since EPI implementation, the epidemiology of measles has changed greatly. For example, over the past 10 years, data show that for children younger than 8 months, who are ineligible for vaccination, the relative burden of measles has increased. Specifically, although the number of annual measles cases in China generally decreased during 2005 to 2012 (123,136, 99,602, 109,023, 131,441, 52,461, 38,159, 9943, and 6183 respectively), the percentage of annual cases that occurred in Chinese infants aged <8 months increased from 11.3% to 24.5% [8]. In the initial EPI schedule, the first dose, a monovalent attenuated vaccine, was given at 8 months, and the second dose, either a monovalent vaccine, measles-rubella/MR vaccine, or measles-mumps-rubella/MMR vaccine, was administered at 7 years. In 2006, the schedule was revised so that the second dose is administered at 18–24 months.

Since 2000, annual measles incidence has remained below 10 per 100,000. Between 2005 and October 2013, a total of 596,391 measles cases and 368 measles-related deaths were reported in China, and annual incidence, in cases per 100,000 population, decreased from 9.95 in 2008 to 0.46 in 2012 but then rose to more than 1.96 in 2013 [6]. The reason for this increase under the EPI may be explained by waning maternal antibody protection, which for most infants is likely derived from maternal vaccination rather than natural infection [5,9–12].

Hand foot and mouth disease (HFMD) is a common infectious disorder caused by enterovirus. In the western Pacific region, two enteroviruses, human enterovirus 71 (EV71) and coxsackievirus A16 (CoxA16), co-circulate and are the principal agents of HFMD. Some patients with EV71 infection rapidly develop neurological and systemic complications

that can be fatal [13,14]. In China 7,200,092 probable cases of HFMD were reported to the national surveillance system during 2008–2012, of which 2457 (0.03%) were fatal [6]. Two alum-adjuvant inactivated EV71 vaccines developed in mainland China, which showed high efficacy (94.8% against disease, 100% against EV71-associated hospitalization or neurologic complications), good immunogenicity persistence and acceptable safety profiles in clinical trials, have been approved for HFMD prevention among Chinese infants [15,16]. The vaccine, which is initially administered at 6 months, has been licensed in China since December 2015 [16,17]. The current vaccination schedules for both measles and EV71, as well as the absence of a vaccine for CoxA16, permit assessment of how waning maternal antibody protection is affecting rates of infant infection in China.

The effects of waning maternal measles antibodies and very early susceptibility to measles among infants have been well studied in countries where measles has been eliminated. For example, in Belgium, geometric mean titres (GMT) of IgG against measles (enzyme linked immunosorbent assay; ELISA) and proportions of positive samples decreased monotonically from 1593 mIU/ml (181/214) for women at week 36 of pregnancy, to 1369 mIU/ml (152/189) for cord blood, 928 mIU/ml (122/160) for infants at 1 month, 304 mIU/ml (72/158) at 3 months, 79 mIU/ml (11/72) 6 months, and 11 mIU/ml (0/156) at 12 months [5]. Similarly, in Taiwan, a cohort study was conducted to understand the dynamics of maternal EV71 antibodies in infants at 0 and 6 months [18]; however, no study has been specifically designed to reveal the antibody dynamics in infants for CoxA16, which typically produce less severe complications than EV71.

In China, prior studies examining measles, EV71 and CoxA16 antibodies among different infant age-groups mostly employed cross-sectional designs [9,17,19–21], which cannot reveal the ‘true’ evolution of antibody levels. Using a longitudinal study design, we here aim to report how maternally-derived antibodies in infants for measles, EV71 and CoxA16 evolve over time in infants during the first 6 months post-partum prior to vaccination.

2. Methods

Guangzhou is the economic, educational and cultural hub of southern China. The city has a sub-tropical climate, a permanent population of 7.94 million residents, a transient population of 4.67 million, and approximately 60 000 births each year in 2010. With 45 beds and nearly 2500 children delivered per year, the Liwan District Maternal and Child Health Hospital provides medical services for pregnant women living in the area, as well as some women from nearby Southsea county. During July 2013 to April 2014, infants born at the Liwan District Maternal and Child Health Hospital in Guangzhou, southern China were enrolled for this study by convenience sampling. We included healthy pregnant women and their healthy offspring. Cord blood from newborns, and venous blood at 3 months and 6 months of age were collected. Inclusion criteria were age 16–45 years and residence in Guangzhou or the Southsea county in Guangdong province, southern China for the duration of the study. Exclusion criteria were the presence of an immunodepressive condition or an acute infection, or administration of immunoglobulins or blood products during the study period. Maternal demographic and health information including age, education, hypertension, diabetes, anemia, and gestation week before delivery, as well as the corresponding newborn

gender, birth weight and Apgar Score after birth, were collected by questionnaire by the study nurses. In China, no specific infectious disease reporting or vaccination records system existed two decades ago. In lieu of these data, we attempted to collect information regarding prior infection and vaccination from study participants; however, most of the participants found it difficult to recall their infection or vaccination history. As a consequence, we did not include this information in our analysis.

Several tests were employed to assess antibody levels in the collected serum samples: (1) the ELISA (Anti-Measles IgG and IgM, Virion/Serion, Germany) [20] was used to quantify measles antibody concentration; and (2) a modified cytopathogenic effect assay [22] was performed to evaluate neutralizing antibody titers against coxA16 (CoxA16; G10 strain, 7.0 lgCCID50/ml) and EV71 (H07 strain, 7.3 lgCCID50/ml [22]. Equivocal assay results were retested once.

Protective antibody cut-off values of 200 mIU/ml for measles were used in this analysis, per prior work [20,23]. WHO Western Pacific Region Regional Reference Measles Laboratory tested a panel of sera and found that the enzyme immunoassay (Vrion/Serion kit) with a cut-off of 200 mIU/ml, when compared to the Plaque Reduction Neutralization Test (PRNT), showed a sensitivity of 94.9% and a specificity of 100% [24]. Consequently, the ELISA provides a conservative measure for elevated measles antibody levels.

Positive cut-off values of 1:8 for CoxA16 and EV71 were also employed [22]. GMT and 95% confidence intervals (CI) were calculated to compare the neutralization antibody levels for EV71 and CoxA16. The occurrence of disease symptoms for measles or HFMD, e.g. rash or fever, were recorded by the study nurse during the 3-month and 6-month collections. If rash or fever were observed at 3 or 6 months of age by the guardians, the infants were immediately referred for clinical management and venous blood was collected and tested for measles-IgM and IgG antibodies and neutralizing antibodies titers against EV71 and CoxA16. Infection for EV71 or CoxA16 virus was defined as a 4-fold increase of neutralization antibody level [25].

We used a linear mixed model (LMM) to describe antibody levels (measles, EV71 and CoxA16) in infants over time while accounting for heterogeneity among and homogeneity within infants [26]. To facilitate analysis of antibody waning rates as a function of initial titre, antibody levels at birth were categorized as high (≥3200 mIU/ml), medium (800–3200 mIU/ml) or low (<800 mIU/ml) for measles [20]; and high (≥1:48) or low (<1:48) for EV71 and CoxA16 antibodies [22]. The possibility that different subjects might have different intercepts defining their antibody waning trajectories, in addition to different slopes (representing the relationship of antibody level with month of age) was considered [26]. Fixed effects associated with antibody level, month of age, and the interaction between antibody level and age were also included.

Data analysis was conducted using SPSS statistical software (Version 23.0, SPSS, Inc.). A repeated measure test (GLM) was employed to assess the trend of antibody levels for measles, EV71 and CoxA16 (GMT) at 0, 3 and 6 months. For all analyses, P values less than 0.05 were regarded as significant. The study protocol was reviewed and approved by the

Guangzhou Center for Disease Control and Prevention Ethics Board and written consent was obtained from all guardians (ClinicalTrials.gov Identifier: NCT02219061).

3. Results

Seven hundred and seventeen newborns were enrolled for this study. Of 715 pregnant women, three had twins and 36.4% (260) gave birth by caesarean. 71.2% (509) had not attended college; 64.8% (463) lived in a family with 2 or 3 persons, and 35.2% (252) with ≥4 persons. Fifty-three percent of the newborns (380/717) were male. General characteristics of the enrolled subjects are provided in Table 1, which shows an overlap of the 95% CIs for the baseline characteristics and antibodies levels for all subjects at 0, 3 and 6 months.

We assessed measles IgG antibody concentration from 717, 233 and 75 sera in infants of 0 month, 3 months and 6 months and 225, 217, and 72 sera for EV71 and CoxA16, respectively. Waning of maternal antibodies for measles, EV71 and CoxA16 was observed during the first 6 months of age. For measles, there was a monotonic decrease of antibody concentration from month 0 (1410 mIU/ml), to month 3 (195 mIU/ml), and month 6 (22 mIU/ml) (Test of Sphericity, $p < 0.001$; $F = 60.759$, $p < 0.001$) (Fig. 1). A similar significant trend was observed for EV71 (1:19.9, 6.3 and 4.5) ($F = 54.765$, $p < 0.001$) and CoxA16 (1:16.3, 5.9, and 4.5) ($F = 65.578$, $p < 0.001$) (Fig. 2). Based on these assays, 25.3% (95% CI, 20.1–31.2) infants 3 months old and 2.7% (95%CI, 0.6–8.3) 6 months old were measles antibody positive. The percentage of infants antibody seropositive for EV71 and CoxA16, respectively, was 72.2% (95%CI, 66.2–77.8) and 72.7% (95%CI, 66.6–78.2) at 0 months, 33.2% (95%CI, 27.2–39.6) and 30.4% (95%CI, 24.6–36.8) at 3 months, and 6.8% (95%CI, 2.7–14.4) and 5.6% (95%CI, 1.9–12.7) at 6 months. When we restricted the analysis to only those sera obtained from infants at all three time points, similar trends for the three antibodies were observed (Table 2).

LMM findings indicated that infants with higher antibody titers at birth experienced a greater depletion of antibody levels. Specifically, infants with high measles antibody levels (≥3200 mIU/ml) at birth (mean, 4512 mIU/ml) waned by 2552 mIU/ml every 3 months versus 1028 mIU/ml every 3 months for those with medium levels (800–3200 mIU/ml) at birth (mean, 1716 mIU/ml), and 246 mIU/ml for those with low levels (<800 mIU/ml) (mean, 366 mIU/ml at birth) ($F = 356.6$, $p < 0.001$). Similarly, for EV71 and CoxA16, infants born with titers ≥ 1:48 waned by 5.7 and 4.8 every 3 months compared to 1.6 and 1.7 for titers < 1:48 at birth ($F = 268.2$ and 175.5, $p < 0.001$). Despite these different antibody depletion rates, infants born with higher measles antibody levels maintained greater maternal protection. The average measles antibody loss as a percentage of original concentration was 56.6% (2552/4512), 59.9% (1028/1716), and 67.2% (246/366) for the categories ≥3200, 800–3200 and <800 mIU/ml at birth, respectively. However, the waning of EV71 or CoxA 16 antibody titers were equivalent, regardless of antibody levels at birth (see Tables 3 and 4).

No relevant symptoms (such as rash or fever) for measles or HFMD were identified among study subjects, and all infants were measles-IgM negative. However, one infant’s CoxA16

neutralizing antibody titer increased from 1:4 at month 0 to >1:1024 at month 6; and one infant's EV71 antibody increased from 1:24 at month 0 and month 3 to 1:256 at month 6, indicating an infection rates of 1.33% (95%CI, 0.1–6.1) for both EV71 and CoxA16.

4. Discussion

This study is among the first to present the evolution of measles and EV71 antibody levels among newborns in China, and is also the first to report the specific dynamics of CoxA16 antibodies. Using pre-vaccination longitudinal data at 0, 3 and 6 months, we find considerable waning of maternal measles IgG antibody (1410, 195, and 22 mIU/ml), and neutralizing antibodies against EV71 (1:19.9, 6.3 and 4.5) and CoxA16 (1:16.3, 5.9, and 4.5). For subjects at 6 months of age, 2.7% (95%CI, 0.6–8.3), 6.8% (95%CI, 2.7–14.4) and 5.6% (95%CI, 1.9–12.7) were antibody positive for measles, EV71 and CoxA16, respectively. Similar trends are observed when the analysis is restricted to infants contributing sera samples at all three times points. LMM findings indicate that infants with higher antibody levels at birth experienced a more substantial depletion of those antibodies.

This study provides solid evidence of the rapid waning of measles antibodies in infants prior to vaccination under the EPI schedule in China. It also confirms the rapid decrease of measles antibody levels suggested by prior cross-sectional studies in China. Compared with our findings of 87.4%, 25.3%, and 2.7% sero-positivity among infants at 0, 3 and 6 months, Xu et al. reported overall sero-positivity of 89.3% (month 0), 22.3% (month 3), and 6.9% (month 6) in three cities in eastern China using the same assay kit; the study also documented sero-positivity at 6 months of 14.9%, 4.1% and 1.6% in Jinan, Ningbo, and Harbin City, respectively [21]. Employing a different ELISA kit, manufactured by the China Center for Disease Control and Prevention, Zhang et al. found a decreased sero-positivity rate in Qinghai, the poorest province in China, of 98.0% (95%CI, 87.4–99.7) at month 0, 91.8% (80.2–96.9) at month 3 and 68% (54.0–79.4) at month 6 in 2009 [21]. In Qinghai province, measles has remained endemic since the establishment of EPI with quadrennial epidemic cycles, and the average incidence of measles during the 1990s was 33 cases per 100,000 [7]. These circumstances may be indicative of a higher prior infection rate, low vaccination coverage, and population groups unvaccinated or partially vaccinated, all of which could contribute to continual measles outbreaks. As a result, more maternal antibodies derived from natural infection and higher infant antibody levels may be induced. Unlike the Qinghai province, most areas in China have experienced low measles incidence records in recent years [8].

The present study found a sero-positivity rate of 2.7% at 6 months of age in Guangzhou, China; however, in a previous cross-sectional study, we found a sero-positivity rate for measles antibody of 48.5% (95%CI, 41.7–55.4) among infants 5–7 months of age in Guangzhou city [9]. The difference between the former and present study is unclear, but may stem from discrepancies in sampling approach, the ELISA kit employed, or the study year.

By using longitudinal data, we were also able to report depletion rates among groups with different measles antibody levels at birth. Infants born with higher titers tend to have more antibody loss than those born with lower titers. The mechanism for this difference is unclear

and further study may be needed [4]. On the other hand, although newborns with higher measles antibody levels usually experienced more antibody loss, their titer levels remained higher than infants born with lower antibody levels.

Due to waning maternally-derived measles antibody levels, young infants are at greater risk for measles. Indeed, in a city in southern China, from 2005 to 2014, the proportion of total measles cases among children <1 year increased from 24.3% to 47.9% [27]. In Tianjin, northern China during 2014, 558 measles cases (20.58%) were among infants aged <1 year [28].

As a consequence, strategies for measles vaccination among young infants or women of childbearing age may need to be reconsidered in China. For young infants, it is critical that the optimal age for measles vaccination be identified that balances the risk from maternal antibody loss with the risk of primary vaccine failure due to the presence of maternal antibodies [29,30]. Indeed, it has been shown that measles vaccination at an early age may not be effective, as maternal antibodies in young infants may impede vaccine protection by disrupting vaccine-induced immune responses [31]. Such interference has generally motivated against early measles vaccination. Alternatively, if more infants are born with higher antibody levels, the risk of infection between 0 and 8 months might be reduced. High maternal antibody levels in pregnant women are vital for efficiently transferring these antibodies to the newborn [32].

For this study, we did not have the opportunity to measure measles antibody levels at 8 months of age and determine explicitly the possible benefit of vaccination 2 months earlier at 6 months of age. We obtained the sera from infants ≤6 months of age in the study hospital where parents take their infants for regular medical examination; however, children >6 months age seldom go to the hospital, which thus precluded explicit assessment of the additional antibody level loss from 6 months to 8 months. Future measurement of this difference would more definitively determine the optimal age for initial measles vaccination.

This longitudinal study reveals how EV71 antibody levels wane in infants during the first half year of life. In Taiwan, Luo et al. reported that maternal EV71 neutralizing antibodies declined from 50% in neonates to undetectable levels in 99% of children at 6-months [18]. A retrospective study conducted in Jiangsu Province in eastern China showed that antibody seropositive rates against EV71 among infants aged 2 and 7 months were 57.6% (95%CI, 54.5–60.8) and 39.5% (95%CI, 36.4–42.6), respectively [22]. The rates from this latter study are higher than what we report here. Further, in the first 6 months post-partum, the HFMD incidence rate of 0.72% (7/975) from EV71 in Jiangsu study was lower than our findings (1.33%), albeit with a smaller sample size. These differences could stem from different infection rates among the populations in southern and eastern China [33].

The immunogenicity of the EV71 vaccine (strain H07, subgenotype C4; 400 U of EV71 antigen with alum adjuvant) administered at 6 months has been noted in randomized trials in China [16,34]. Although the incidence for EV71 infection was very low in infants ≤6 months of age, the case-fatality risk, case-severity risk, and severity-fatality risk have been reported much higher than for other age groups [6]. Given the decline of maternal

neutralizing antibodies, this date of immunization appears to be too late, at least for the Guangzhou population, to provide protection to infants ≤6 months who more commonly incur severe outcomes from EV71 infection [6].

Our findings show that 72.7% of newborns, 30.4% of infants at 3 months, and 5.6% of infants at 6 months are protected from CoxA16 infection. These levels are lower than reported previously in the Jiangsu retrospective study (41.3% and 26.4% at months 2 and 7, respectively) [22]. Although CoxA16 infection usually causes mild symptoms, severe and fatal HFMD cases due to CoxA16 infection have been reported in the United States, France, and Japan [35–37]. In Shenyang, northeast China, 20.7% (19/92) of HFMD cases presenting with neurological symptoms were due to CoxA16 infection, with 2 patients presenting with brainstem encephalitis and one with acute flaccid paralysis[38]. Additionally, studies indicate that co-infection with CoxA16 and EV71 viruses might increase the possibility of genetic recombination between the two viruses [39–41]. Given the dramatic waning of maternal immunity and possible severe clinical outcomes from CoxA16 infection[42], it is advised that development of human CoxA16 vaccine (monovalent, or bivalent EV71 and CoxA16) should be a priority.

There are several limitations in this study. The first is the high drop out rate – i.e, 85% of subjects did not provide a sample at the third point. However, the general demography and initial antibody levels among subjects providing 3 samples are comparable to the overall cohort at month 0. The second possible limitation is that the subjects from the one hospital we used for sampling may not be representative of the overall population. However, some of subject characteristics, such as women's education level (71.2% who had not attended college) or gender ratio of newborns (53% were male) in the sample are similar to the general demographics in Guangzhou (76.3% and 51.2% respectively) [43]. Finally, we failed to collect information on measles infection or vaccination from the pregnant mothers, and thus we cannot compare the antibody levels and wane rates between these two groups. These limitations suggest that additional investigations are needed before applying these findings broadly in China to inform vaccination policy.

In sum, our findings demonstrate a waning of maternally-derived antibodies in infants prior to vaccination for measles, EV71 and CoxA16. Further modifications of vaccination strategies for measles, earlier vaccination for EV71 infection, and development and provision of a CoxA16 vaccine should be investigated and considered in the future.

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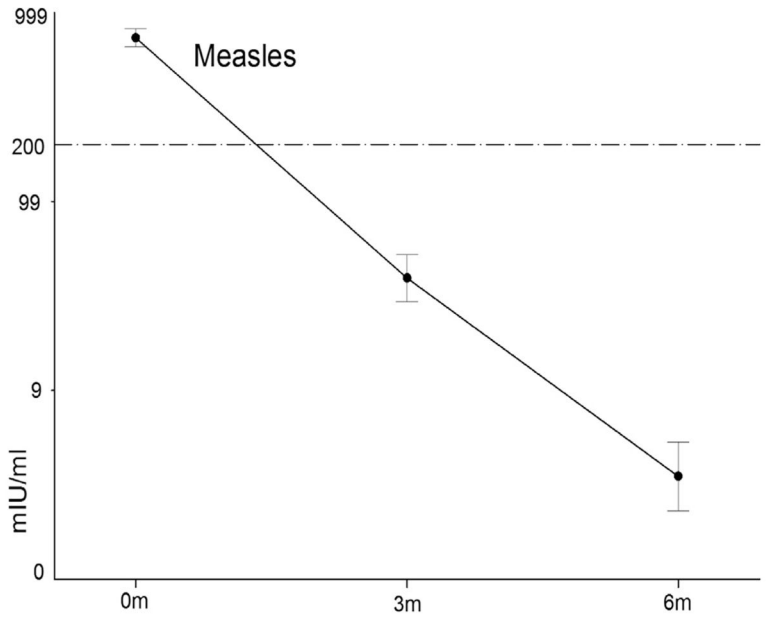

Fig. 1.

Profile for the decay in log measles antibodies based all sera at 0, 3 and 6 months. Log (concentration + 1) was computed for measles antibody. The mean and 95% confidence intervals are shown. All infants tested measles-IgM negative and no cases of measles infection were identified. No. of sera: 717, 233, 75 at 0, 3 and 6 months of age.

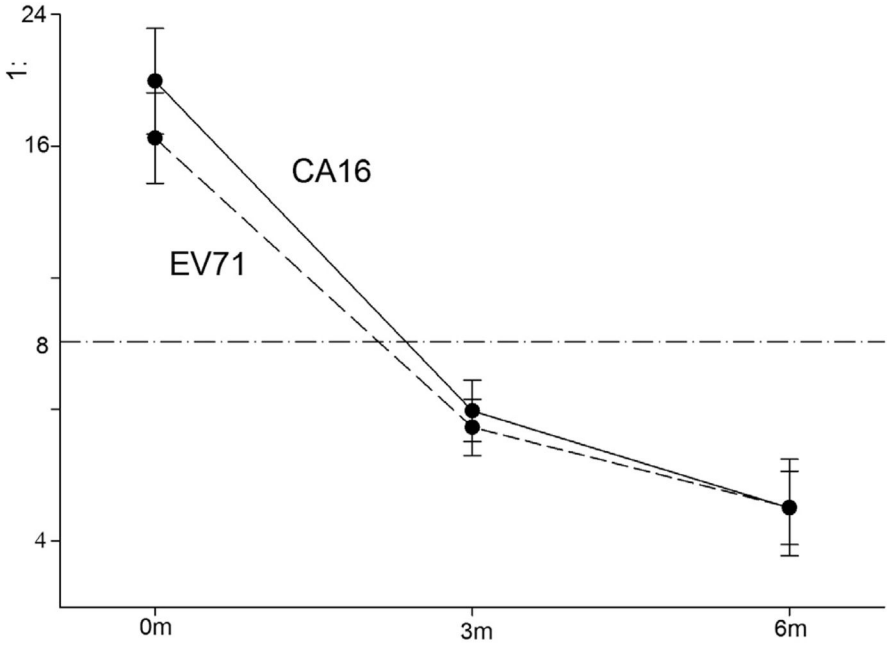

Fig. 2.

Profiles for the decay in log antibodies based all sera at 0, 3 and 6 months for coxsackievirus 16 and enterovirus 71. Note. The number of sera was 225, 217 and 72 at 0, 3 and 6 months.

Table 1

Baseline characteristics and antibody levels (mean value) for all subjects of 0, 3, 6 months of age. Changes in maternal age, gestational age, birth weight, and Apgar Score reflect differing sample sizes.

	0 mo (N = 717)	**3 mo (N = 233)**	**6 mo (N = 75)**
Mean (min, max)			
Mother age (years)	27 (16,45)	28 (18,42)	28 (20,42)
Gestational age (weeks)	39 (32,43)	39 (35,41)	39 (36,41)
Birth weight (kg)	3.19 (1.38, 4.82)	3.24(2.10,4.44)	3.22 (2.35, 4.04)
Apgar score 1-min	9 (6,10)	9 (7,10)	9 (8,10)
Apgar score 5-min	10 (8,19)	9 (8,10)	10 (10,10)
Means with 95% CIs			
Measles (mIU/ml)	1409.9 (1305.8,1514.5)	1192.4 (1029.5,1363.9)	1631.6 (1284.3,1995.1)
Enterovirus 71 (1:)	19.9 (16.6, 24.0)	19.6 (16.1, 23.9)	20.5 (14.4, 29.0)
Coxsackievirus A16 (1:)	16.3 (13.9, 19, 1)	16.6 (14.0, 19.7)	16.2 (12.3, 21.4)

Differences in maternal age, gestational age, birth weight, and Apgar score (1 and 5 min) between infants at 0 month and 3 months of age, or between infants at 0 month and 6 months of age were non-significant.

Table 2

Measles, Enterovirus 71 and coxsackievirus 16 antibody GMT at 0, 3 and 6 months of age, based on the paired sera.

	N	Mo 0	Mo 3	Mo 6
Measles	56	1482.6 (1103.8, 1861.3)	253.4 (152.9, 353.9)	20.0 (4.7, 35.3)
CoxA16	51	17.4 (12.5, 24.3)	6.1 (4.8, 7.7)	4.2 (4.0, 4.4)
EV71	52	19.7 (13.0, 29.7)	6.4 (5.2, 7.9)	4.2 (4.0, 4.4)

Table 3

Mean measles IgG antibody concentrations (mIU/ml) at 0, 3 and 6 months of age for the 3 initial antibody level categories. 95% confidence intervals are given in parentheses and sample size is in brackets.

Initial antibody category	Mo 0	Mo 3	Mo 6
≥3200 (N)	4512.1 (4190.6, 4833.7) [78]	905.3 (408.9, 1401.6) [21]	91.6 (13.7, 169.6) [10]
Proportion	100%	20.0%	2.0%
800–3200(N)	1716.0 (1642.7, 1789.3) [315]	240.7 (190.2, 291.2) [92]	15.5 (6.3, 24.7) [39]
Proportion	100%	14.0%	0.9%
<800(N)	365.5 (340.6, 390.4) [324]	35.0 (20.9,49.2) [119]	4.0 (−4.0,12.2) [26]
Proportion	100%	9.6%	1.1%
Total	1409.9 (1305.8,1514.5)	194.6 (138.5,250.6)	21.7 (9.5, 33.8)

The initial antibody level (Mo 0) was taken as 100% and the proportion for measles antibody were shown for Mo 3 and Mo 6.

Table 4

Enterovirus 71 and coxsackievirus 16 antibody GMT at 0, 3 and 6 months of age for the 2 initial antibody level categories. 95% confidence intervals are given in parentheses and sample size is in brackets.

	Initial antibody category	Mo 0	Mo 3	Mo 6
EV71(N)	≥1:48	115.2 (96.1, 138.1) [71]	13.3 (10.5, 16.9) [63]	4.7 (3.9, 5.8) [22]
	Proportion	100%	11.5%	4.1%
	<1:48	8.9 (7.9, 10.1) [156]	4.4 (4.2,4.6) [138]	4.4 (3.7, 5.3) [47]
	Proportion	100%	49.4%	49.4%
CoxA16(N)	≥1:48	83.9 (67.2,104.9) [55]	14.3 (10.9,18.9) [50]	4.5 (3.8, 5.4) [15]
	Proportion	100%	17.0%	5.4%
	<1:48	9.7 (8.6, 10.8) [172]	4.5 (4.3,4.8) [151]	4.5 (3.6,5.7) [53]
	Proportion	100%	46.4%	46.4%

The initial antibody level (Mo 0) was taken as 100% and the proportion for EV71 and CoxA16 antibodies were shown for Mo 3 and Mo 6.