Brief Reports

Immunogenecity of Edmonston—Zagreb Measles Vaccine

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Measles in infants under 9 months of age is recognized as a major health problem in some parts of the world(l). In areas, where intense transmission of measles occurs, infants may be exposed to the virus and develop measles as soon as their maternal antibody wanes to non protective levels(2). Previously, it was thought that increasing coverage in older children would decrease transmission to younger infants. However, recent reports from urban areas in Africa document that a large percentage of cases occur in infants less than 9 months of age despite coverage as high as 80% in 12-23 month olds(3). Moreover, measles before 9 months of age is doubly dangerous because of the high mortality afterwards due to a higher incidence of complications(4). In order to offer increasing protection to young infants, WHO proposed several strategies including use of different vaccine strains(E-Z) or doses which may be more immunogenic(5). In literature, there is mounting evidence in favor of Edmonston-Zagreb (E-Z) measles vaccine as apart from being more immunogenic, its immunogenicity is not interfered with by the presence of maternal antibody even in the younger age(6). The present study was undertaken to evaluate the immunogenicity and safety of E-Z measles vaccine manufactured in India (Serum Institute of India, Pune) in 4-6 month old infants and compare it with subjects aged 9-10 month.

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Subjects and Methods

The study was conducted in the Department of Pediatrics, Indira Gandhi Medical College, Shimla, between October 1992 to November, 1993. Healthy infants attending the Well Baby Clinic and without any history of exposure to measles in the preceding two weeks were randomly enrolled and categorized as Group A (4-6 mo) and Group B (9-10 mo). A detailed clinical examination was carried out to exclude any illness. Infants were given 0.5 ml of (E-Z) measles vaccine on the lateral aspect of thigh subcutaneously. Potency of vaccine used was checked at C.R.I. Kasauli on two occasions during the study, Titers of 4.28 log $_{10}$TCID$_{50}$ and 4.48 Ig$_{10}$ TCID$_{50}$ per dose were obtained. It also passed the sterility test, abnormal toxicity test and identity test on both occasions. Prevaccination blood samples were collected by finger/heel prick on a rectangular No. 1. Whatman filter paper strip inscribed with 14 mm disc, immediately dried, stored in an air tight container at 4-8°C till analysis at C.R.I. Kasauli by Passive Hemagglutination (PHA) test(7). Two post vaccination samples were collected after 6+1 and 24+1 weeks of vaccination in both the groups in the manner described above. Parents were advised to record any side effects following vaccination on the proforma provided and report after two weeks or when the need arose. A PHA titer of 1:8 or higher was taken as seropositive and seroconversion was defined as: *(i)*

two or more fold rise in post-immunization titer, *(ii)* rise of less than 1:8 titer to 1:8 or higher(8). Data thus collected was analyzed on Computer and standard statistical packages were used to calculate paired Student 't' test and chisquare tests. Geometric mean titer (GMT) was calculated only for seropositive infants.

Results

Initially 200 infants (100 each in Groups A and B) were enrolled but only 163 (81 in Group A and 82 in Group B) completed the study. There was no sex wise difference in results, hence gender was ignored for further analysis. Out of 81 infants in Group A, 11 were four (5 males and 6 females), 24 five (13 males and 11 females) and 46 six mo (24 males and 22 females) old and of the 82 infants in Group B 42 were males and 40 females.

Prevaccination Antibody Status

PHA titer was negative (<1:8) in 43.2% (35/81) infants in Group A and 76.8% (63/82) infants in Group B (P <0.05; *Table I).*

Seroconversion

In Group A, the overall seroconversion at 6 weeks was 65.4% (53/81) and 74.1% (60/81) at 24 weeks (p >0.05) *(Table I).* The rise in GMT from preimmunization was not significant in 4, 5 or 6 mo old subjects. The rise in GMT from 6 to 24 weeks was only significant (p <0.05) in 5 and 6 mo age group. In Group B, overall seroconversion at 6 and 24 weeks was 86.6% (71/82) and 92.7% (76/82), respectively, (p <0.05). Rise in GMT from preimmunization to 6 weeks was insignificant (p >0.10) but from 6 weeks to 24 weeks significant (p <0.001).

On comparing the seroconversion between Groups A and B, the differences at 6 and 24 weeks were significant (p <0.01 and p <0.003, respectively), thereby indicating a comparatively poorer seroconversion at younger age. When only 6 month old infants of Group A were compared with Group B, the difference at 6 weeks was significant (p <0.01) but at 24 weeks insignificant (p >0.05) thus indicating that the ultimate seroconversion in 6 months infants is similar to that in 9-10 month subjects.

Prevaccination Seronegatives

In Group A *(Table II),* 47.1% (33/70) infants were seronegative and 81.8% (27/33) of these showed seroconversion at 6 weeks (62.3% five month and 88.0% six month olds, p >0.10) and 90.9% (30/33) at 24 weeks (87.5% five month and 92.0% 6 month olds, p >0.10). The difference in overall seroconversion at 6 and 24 weeks was not significant (p >0.1). The GMT for five and six month olds at 6 weeks was 43.65 ± 13.94 and 21.38+15.28, respectively and at 24 weeks 51.29+24.09 and 36.31±21.18, respectively and the rise from 6 to 24 weeks was significant for both ages (p <0.01).

In Group B, 63 (76.8%) out of 82 infants were seronegative and 93.2% (59/63) of these seroconverted after 6 weeks and 95.2% (60/63) after 24 weeks, with corresponding GMT of 23.18±10.27 and 28.95+16.67, respectively (p=0.01 for paired 't' test from 6 to 24 weeks). The difference in seroconversion between Groups A and B at 6 as well as 24 weeks was insignificant (p >0.1 and 0.5, respectively).

Prevaccination Seropositive

In Group A, on the whole 59.2% (37/70) infants were seropositive and nineteen of these (51%) (8/16, five month and 11/21 six month olds) seroconverted at 6 weeks and 78.4% (29/37) at 24 weeks (12/16 five month and 17/21 six month olds). The difference between 6 and 24 weeks seroconversion was significant (p=0.02). The GMT at 6 weeks was 20.42±9.59 and

TABLE I– *Pre and Post Immunization Antibody Titers and Seroconversion*

Sl. No.	Group	Age (mo)	No.	Pre–Immunization Titer			Post–Immunization Titer and Sero Conversion			
				Sero Negative	Sero Positive	GMT ±SD	6±1 Weeks		24±1 Weeks	
				<1:8 N (%)	≥1:8 N (%)		N (%)	GMT ± SD	N (%)	GMT ± SD
1	A	4	11	2 (18.2)	9 (81.8)	16.60 ±17.28	5 (45.5)	23.99 ±7.83	6 (54.5)	28.18 ±18.65
		5	24	8 (33.3)	16 (66.6)	15.85 ±15.44	15 (62.5)	23.99 ±7.2	16 (66.7)	35.48 ±19.39
		6	46	25 (54.4)	21 (45.6)	20.42 ±16.80	33 (71.7)	21.88 ±17.59	38 (82.6)	36.31 ±20.02
	Total:		81	35 (43.2)	46 (56.8)	16.79 ±23.39	53 (65.4)	22.96 ±13.87	60 (74.1)	34.43 ±20.06
2	B	9-10	82	63 (76.8)	19 (23.2)	12.59 ±16.28	71 (86.6)	15.49 ±14.41	76 (92.7)	30.90 ±17.24

TABLE II– *Seroconversion in Relation to Prevaccination Antibody Status.*

Sl. No.	Pre-Vaccination Status	Post-Vaccination Interval (Weeks)	Group A						Group B		
			Five Month Olds			Six Month Olds			(9-10 Months)		
			Sero Converstion			Sero Conversion			Sero Conversion		
			N	n (%)	GMT ± SD	N	n (%)	GMT ± SD	N	n (%)	GMT (±SD)
1.	Sero Negative	6±1	8	5 (62.3)	43.65 ±13.94	25	22 (88.0)	21.38 ±15.28	63	59 (93.2)	23.18 ±10.27
		24±1		7 (87.5)	51.29 ±24.09		23 (92.0)	36.31 ±21.18		60 (95.2)	28.59 ±16.67
2.	Sero Positive	6±1	16	8 (50.00)	20.42 ±9.59	21	11 (52.4)	25.55 ±21.26	19	10 (52.7)	23.99 ±19.74
		24±1		12 (75.0)	25.12 ±13.79		17 (80.9)	32.36 ±19.79		17 (89.5)	33.11 ±18.15

25.55±21.26 for 5 and 6 month olds, respectively and at 2 weeks 25.12±13.79 and 32.31±19.79. The rise in GMT from 6 weeks to 24 weeks was insignificant for 5 months olds ($p > 0.05$) but significant for 6 month olds ($p < 0.05$).

In Group B, only 19 (23.1%) out of 82 infants were seropositive and 52.7% (10/19) of these seroconverted at 6 weeks and 89.5% (17/19) at 24 weeks ($p < 0.02$) and the corresponding GMT was 23.99±19.74 and 33.11+18.15, respectively and the rise in GMT from 6 to 24 weeks was significant ($p=0.01$). On comparing Groups A with B, the difference in seroconversion at 6 as well as 24 weeks was insignificant ($p > 0.05$).

Influence of "Blocking Maternal Antibody" on Immunogenecity of E-Z Vaccine

In Group A, the difference in percentage of seroconversion between prevaccination seronegative and seropositive at 6 weeks was significant (81.5% vs 51.3%, $p < 0.01$) but at 24 weeks insignificant (90.9% vs 78.3%; $p > 0.01$), thus indicating that maternal antibody interferes only with early and not ultimate response to E-Z vaccine in the younger age. Similarly, the difference was also significant in Group B at 6 weeks (93.2% in seronegatives vs 52.7% in seropositive, $p < 0.01$) but insignificant at 24 weeks (95.2% vs 89.5%, $p=0.1$), thus suggesting that even in older age, the early immune response is poorer in presence of maternal "antibodies.

No major adverse reactions were observed in either group. On the whole 13.6% mild reactions were seen in Group A (mild fever-6.2%, cough 3.7%, and diarrhea-3.7%) and 18.3% in Group B (fever-8.5%, cough-4.9%, diarrhea-2.4% and local induration-1.2%). None of the infants under study developed measles over one year follow up period.

Discussion

Our observation of decline in maternally derived antibody with advancing age is in keeping with the literature(6,9) and thus justifies WHO's recommendation for lowering the age for measles vaccination in developing countries.

On comparing the seroconversion behavior in the present study with that of others, the observed rate of 65.5% at 4-6 months was similar to earlier reports(10,ll) in the same age group but lower than 95%(9) obtained with high dose E-Z vaccine at 18 weeks age and 90%(6) at age 4-6 months. Poor seroconversion observed by us in Group A as a whole in comparison to Group B was probably due to a significantly higher number of prevaccination seropositives in the former; however, the ultimate seroconversion of 6 months infants of Group A was comparable to Group B.

While studying the influence of maternally derived antibody on the seroconversion behavior, our observation of 81.8% of seroconversion in prevaccination seronegatives in Group A was in concordance with 94% of Tidjani *et al*.(6) and that of 93.2% in Group B similar to 92% reported by Sheikh *et* a/.(12). Our observation of poorer early and not ultimate immune response in the presence of maternal antibody (prevaccination seropositives) has also been reported by others, who have demonstrated a significant negative correlation between preimmunization antibody titer and the height of ultimate titer obtained after immunization(6,9). Whittle(9) has further stressed that the convention of reporting the success of immunization as the percentage of those who seroconvert is of little relevance when maternal antibody is high at the time of vaccination, what matters is the level of antibody at age 9-10 months as by then maternal antibody is no longer present and this measurement reflects the

child's true response to vaccination and his state of immunity. In our study too, we have already documented a definite increase in GMT from 6 to 24 weeks in Group A, as by then, these 4-6 months infants were beyond 9-10 months of age. The safety of E-Z vaccine in younger age observed by us too has been documented earlier(9,12).

In conclusion seroconversion following E-Z measles vaccine was poorer at younger age but that of six month old infants was similar to 9-10 month subjects. Maternal antibody interferes with only early and not ultimate immune response of E-Z measles vaccine in both younger and older age. The vaccine proved equally safe in younger age.

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